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U1S S1318 S1321 S2413 S2415 S2417 S2418

(56) Documents Cited
Life Sciences, Vol 55 (25/26) 2135-2145,(1994) J. Clinic. Invest., Vol 91, 1314-1318 (1993) J. Pharmacol Exp Ther., Vol 258 (3) 992-998 (1991) Mol Pharmacol, Vol 38 (5) 674-680 (1990) Br. J. Pharmacol, Vol 98 79-86 (1991) J. Cardiovasc Pharmacol Vol 11 (2) 222-229 (1988)

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ONLINE: WPI, CLAIMS, CHEMICAL ABSTRACTS

(54) Allosteric effectors at muscarinic receptors

(57) The present invention provides compounds which have an allosteric effect at muscarinic receptors. This novel effect permits the prophylaxis and treatment of conditions associated with muscarinic receptor dysfunction, such as Alzheimer's disease. Preferred compounds include certain strychnine and brucine derivatives, indole derivatives and pentacyclic tetrahydrocarbazoles.

This print takes account of replacement documents submitted after the date of filing to enable the application to comply with the formal requirements of the Patents Rules 1995
At least one drawing originally filed was informal and the print reproduced here is taken from a later filed formal copy.
The claims were filed later than the filing date within the period prescribed by Rule 25(1) of the Patents Rules 1990.

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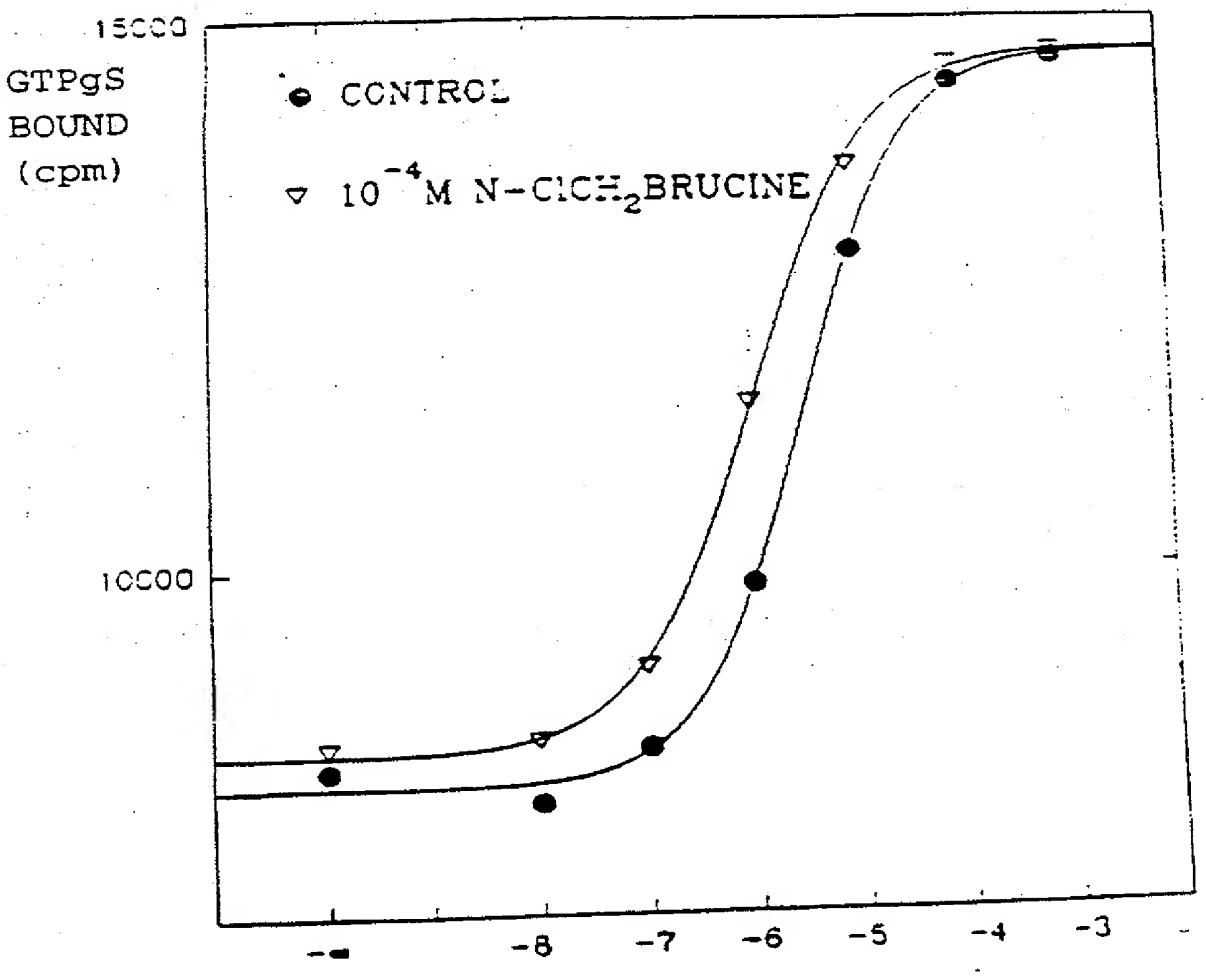
Figure 0

EFFECT OF N-$ClCH_2$-BRUCINE ON ECETYLCHOLINE-STIMULATED [^{35}S]GTPgS BINDING AT m^3



CONTROL $LOG(EC_{50}) = -5.49 \pm 0.05$

N-$ClCH_2$-BRUCINE $LOG(EC_{50}) = -5.95 \pm 0.04$

Figure 1

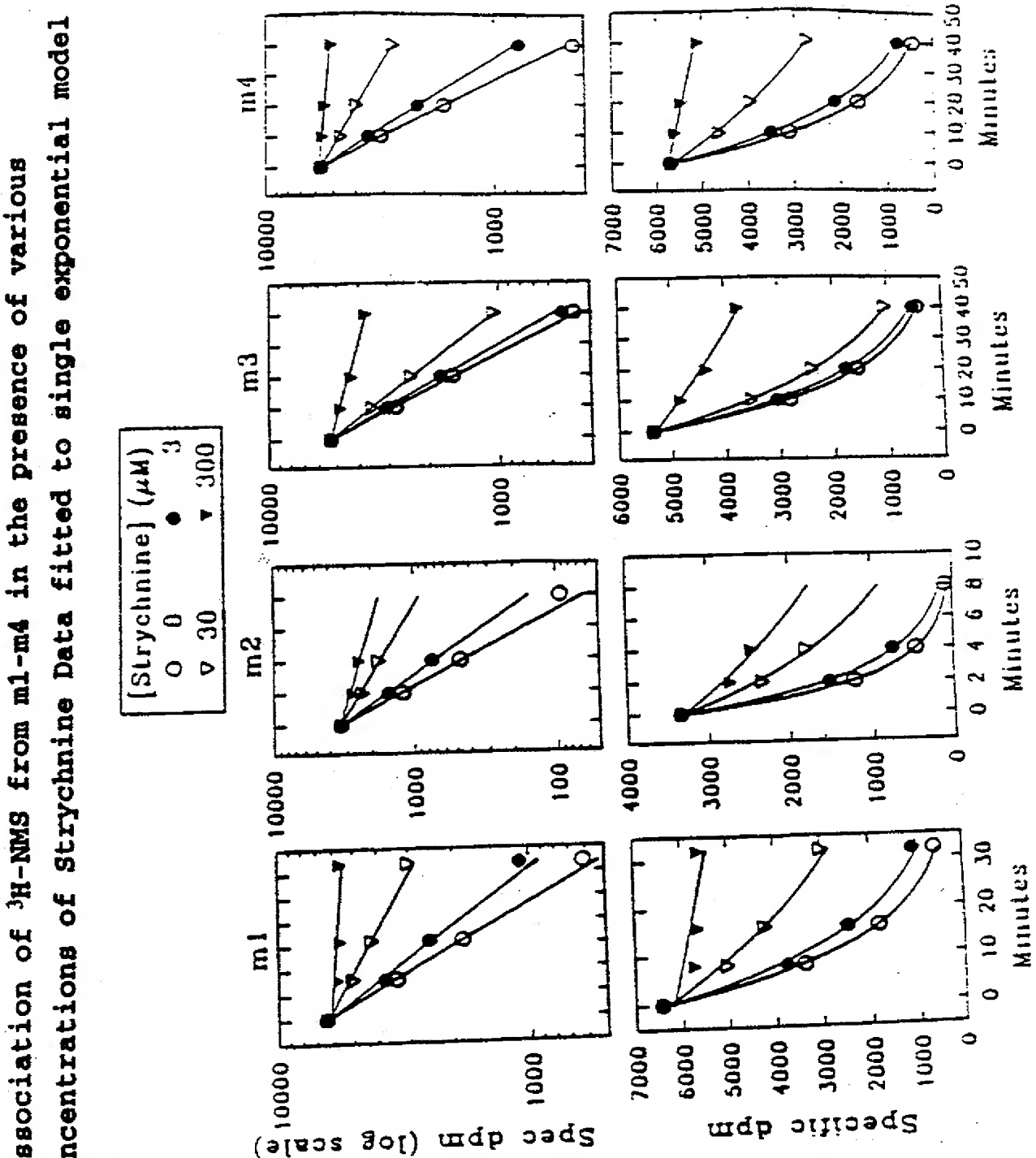


Figure 2

Inhibition of ^{3}H-NMS dissociation rate constant (K_{off} min^{-1}) by strychnine

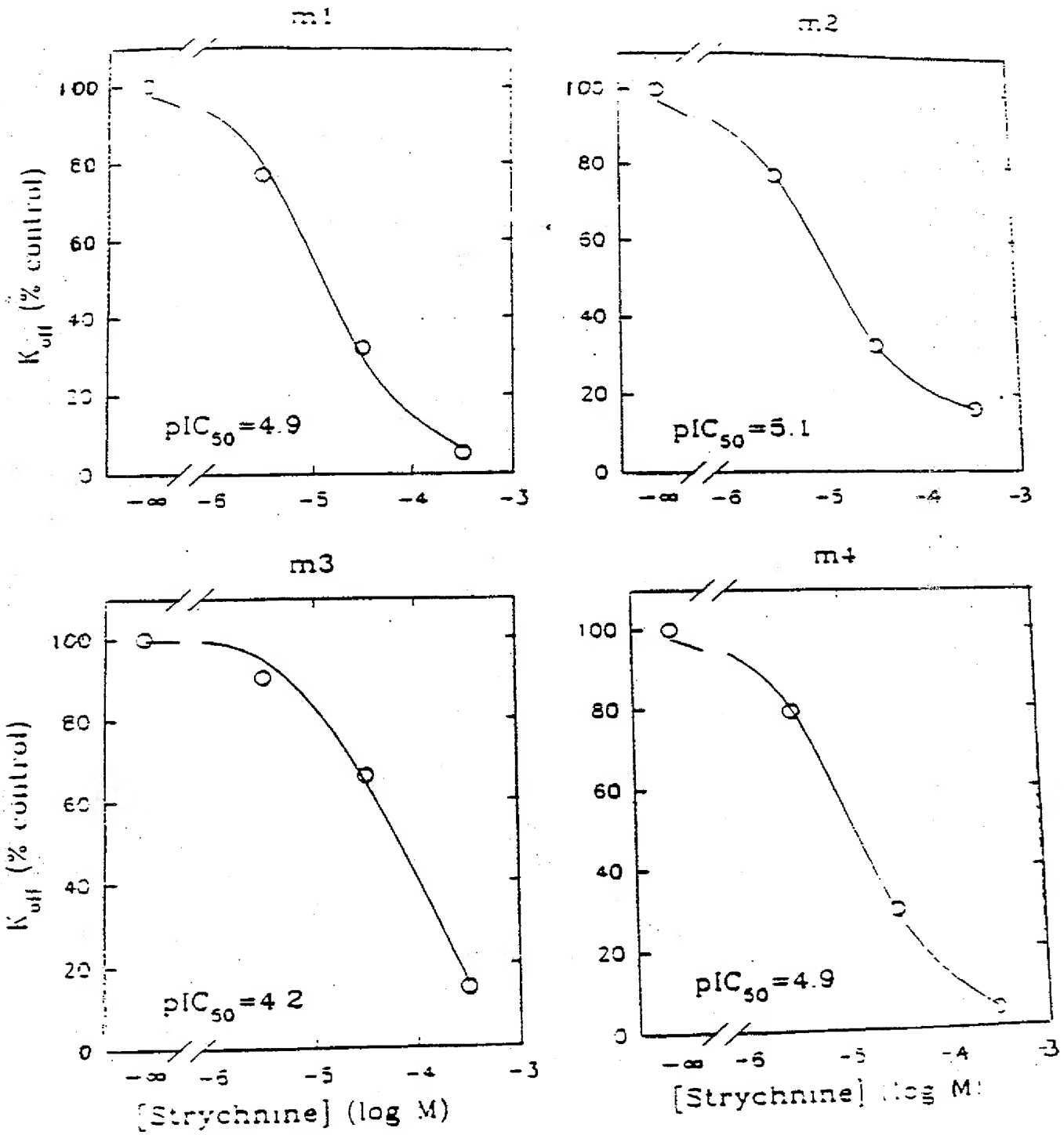


Figure 3

Dissociation of ^{3}H-NMS from m4 in the presence of various concentrations of Strychnine

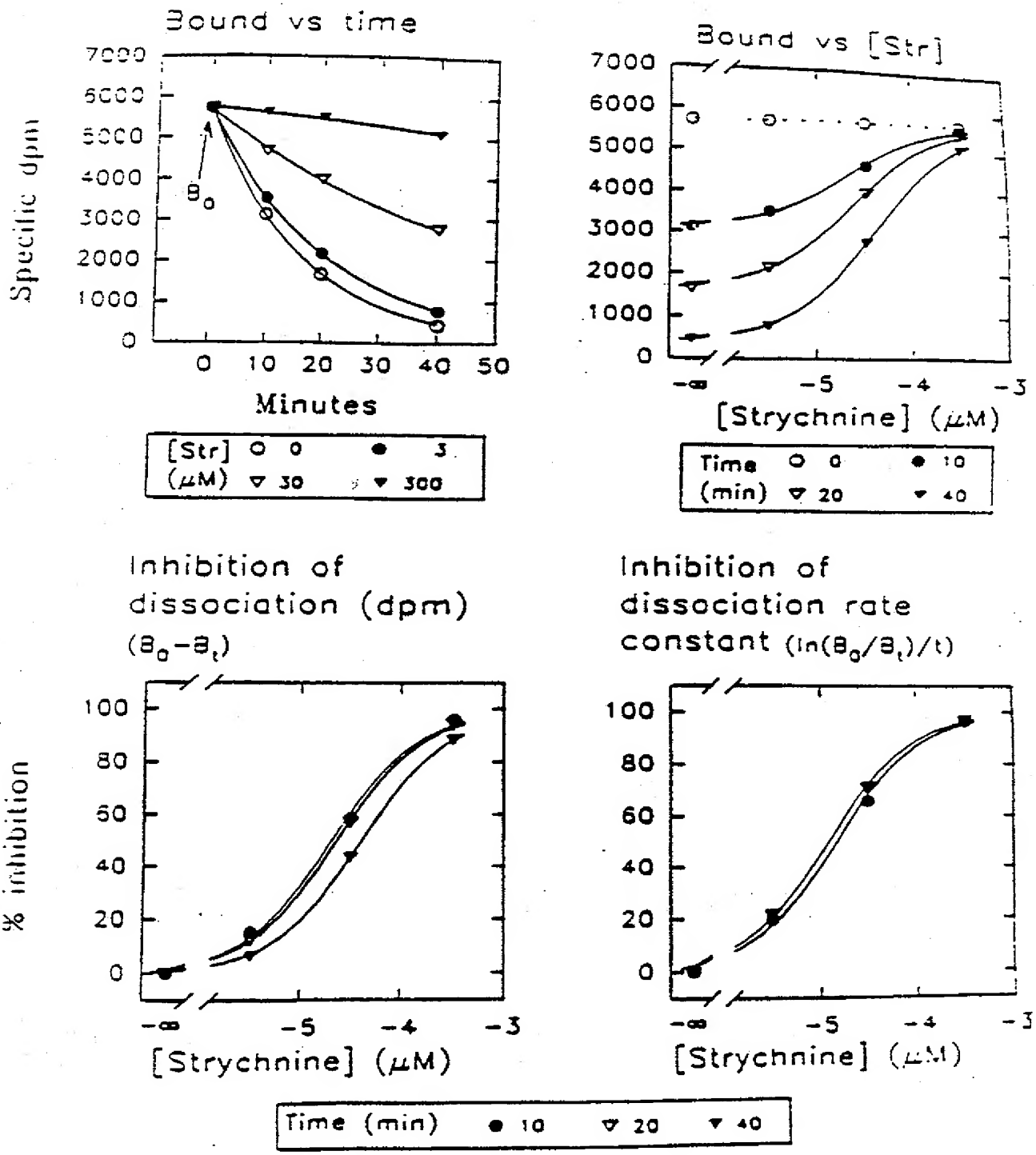
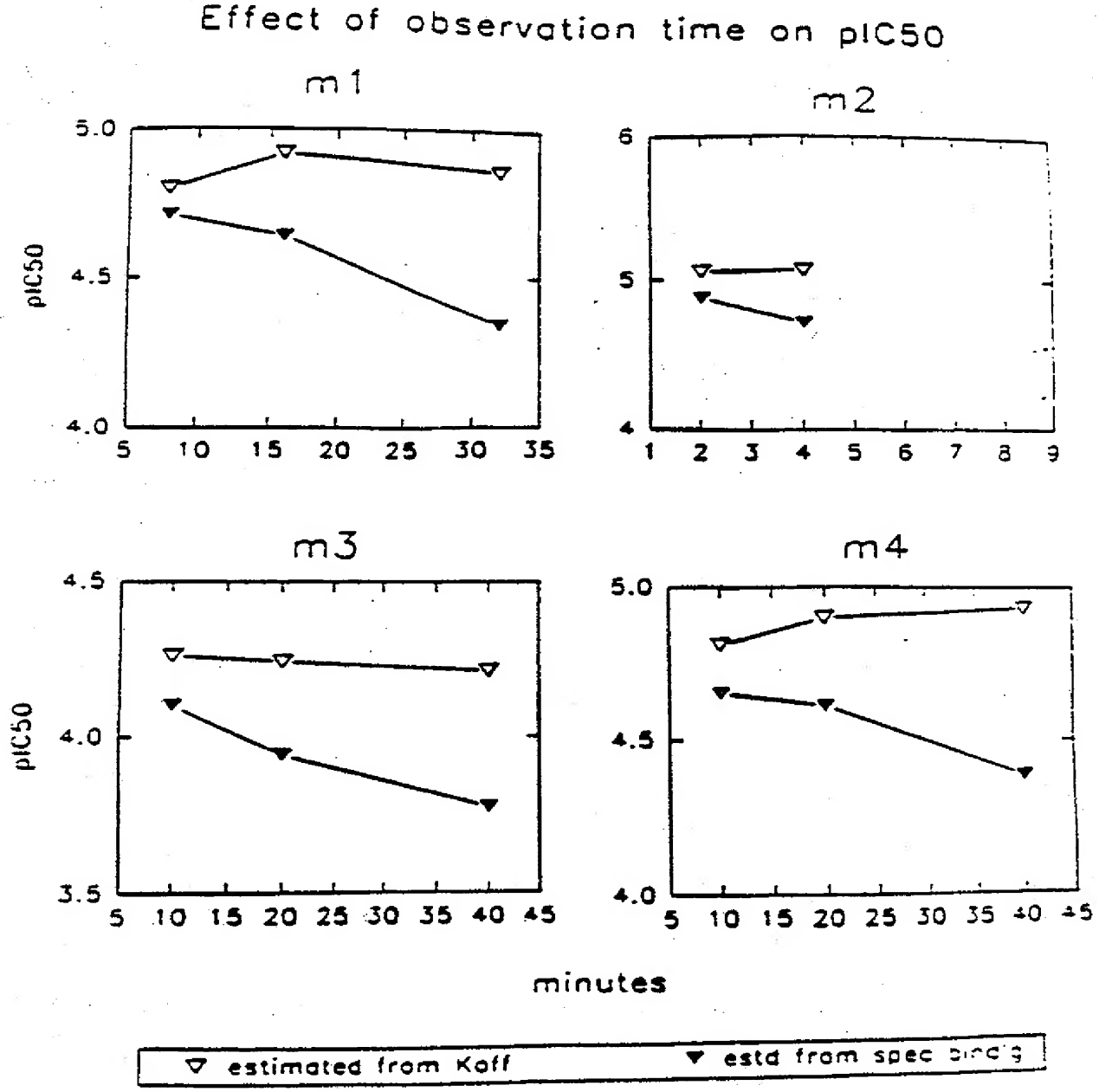


Figure 4

Single-point off-rate screen of Strychnine at m1-m4 measured at 1, 2 and 4 halflives



2292685

ALLOSTERIC EFFECTORS AT MUSCARINIC RECEPTORS

The present invention relates to compounds useful as allosteric effectors at muscarinic receptors, uses of such compounds and synthesis of such compounds.

Acetylcholine is known to be associated with memory, and it is also known that there are decreased levels of acetylcholine in the brain in sufferers of Alzheimer's Disease.

In an attempt to provide a cure or treatment for Alzheimer's Disease, various groups have endeavoured to alleviate the cholinergic deficit in vivo. This has been done, for example, by using cholinesterase inhibitors (to reduce the rate of acetylcholine breakdown) or by using alternative agonists to serve as a supplement to acetylcholine.

Neither course of action has proved successful, as the effect of each is generalised, so that acetylcholine throughout the body and at all receptors is prevented from breaking down, or supplemented (or both), without specifically targetting those receptors involved in Alzheimer's disease. Enhancing the effect of acetylcholine at some receptors can cause depression, for example, so that these courses of action are not being pursued.

More specifically, acetylcholine acts at receptors which fall into two classes; muscarinic and nicotinic. It is believed that the muscarinic receptors are involved in Alzheimer's disease.

The muscarinic receptors belong to the family of

G-protein coupling receptors, and have been classified into three subtypes on the basis of their pharmacological properties and into five subtypes from their molecular structures. The nomenclature of muscarinic receptor subtypes has been confused, and, at the Fourth International Symposium on Muscarinic Receptors, it was recommended that subtypes based on the antagonist binding properties be referred to as M_1, M_2, M_3 and M_4 and that those based on molecular structure be called m1-m5 (see below). This nomenclature is used hereinafter.

Muscarinic receptor nomenclature

Pharmacological characterization					
Subtype	M_1		M_2	M_3	M_4
Selective antagonists	pzpine (+)-tzpne		AF-DX 116, himbacine, m/tramine, gallamine*	p-fluoro-hexahydrosila-difenidol, hexahydrosila-difenidol	tropicamide
Molecular characterization					
Sequences	m1	m2	m3	m4	m5
Numbers of amino acids	460	466	589/590	478/479	531/532

pzpine = pirenzepine; tzpne = telenzepine;
m/tramine = methoctramine; * not competitive;

Recently, it has been possible to use cells expressing m1-m5 receptors. These cells are pure preparations of each receptor subtype and are very useful for characterizing each subtype and screening for subtype specific agents. However, there are few reports which examine agonist and antagonist binding to m1-m5 receptors and their relationship to M_1-M_3 receptors

[Akiba et al., FEBS Lett. 235, 257 (1988); Buckley et al., Molec. Pharmacol. 35, 469 (1989)].

Studies have been performed on muscarinic receptors in the heart (M2) using the antagonist N-methyl-scopolamine (NMS), and these have established that the binding of this antagonist can be affected by other agents, but that these agents do not necessarily act at the NMS binding site. Such action at a different binding site is known as allosteric action, or allosterism. Tucek et al. [J. Neurochem. (1993), 61, Suppl., S19] have shown that the neuromuscular blocking drug alcuronium allosterically increases the affinity of M2 muscarinic receptors in the heart for NMS.

It was reported by Riker and Wescoe in 1951 that gallamine had a negative action on heart receptors [Ann. N. Y. Acad. Sci., 54, 373-94 (1951)]. It was subsequently established that gallamine was not a competitive antagonist for acetylcholine.

Waelbroeck et al. [J. Recep. Res., 8, 787-808 (1988)] reported that curare acts allosterically against muscarinic receptors in the heart, but the allosteric enhancement of ^{3}-H-oxotremorine-M binding cannot be repeated.

Batrachotoxin has also been reported to have negative an allosteric effect on binding.

Birdsall et al. [Pierre Fabre Monograph Series, 1, New Concepts in Alzheimer's Disease, Ed's Briley, M., et al., Macmillan Press, Chapter 9, 103-121] speculate that "the muscarinic receptor sub-types exhibit a selectivity in their binding profile for allosteric agents, and it may hence be possible to selectively 'tune up' muscarinic responses". In this respect, the authors

were referring to the difference between the receptors found in the CNS and those in other parts of the body.

Although not disclosed as allosteric effectors, EP-A-0 171 217 and EP-A-0 415 776 both disclose compounds having a similar structure to those compounds of formula (II) herein.

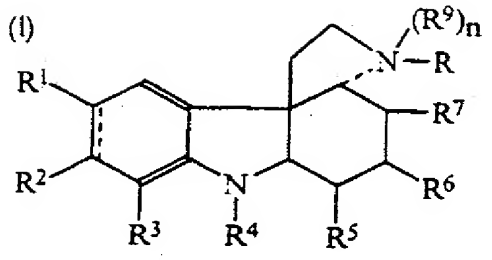
In fact, we have now found that certain compounds are capable of selective activity not only between broad ranges of sub-types, but actually possess different effects even between m1 and m3 receptors, for example. Further, we have identified compounds which have a positive allosteric effect on acetylcholine, including at the m1 receptor.

Thus, the present invention provides, in a first aspect, a method of regulating receptor response in vivo in a mammalian subject, said receptor being selected from m1, m2, m3, m4, m5, M1, M2, M3, M4 and M5 muscarinic receptors, comprising the step of administering to said subject an effective amount of a selective allosteric effector to regulate said receptor.

In the above method, the preferred receptors for regulation are the m1, m2, m3 and m4 receptors, especially the m1 receptor. We prefer that that said effector exhibits positive cooperativity with acetylcholine at said receptor, especially at the m1 recptor.

In an alternative aspect, the present invention provides the use of a compound of formula (I):

Formula (I)



(I)

[wherein the dashed lines independently indicate the presence or absence of a carbon-carbon bond;

R^1 and R^2 are the same or different and, where the dashed line indicates a carbon-carbon bond, may each represent a hydrogen atom, a hydroxy group, a lower alkoxy group, an amino group, an amino group substituted with one or two lower alkyl groups, a nitroso group, a nitro group, a carboxyl group, a carbamoyl group or a carbamoyl group substituted with one or two lower alkyl groups,

or, where the dashed line indicates no bond, then one of R^1 and R^2 represents a keto group and the other represents a keto group or two hydrogen atoms;

R^3 represents a hydrogen atom, a hydroxy group or a lower alkoxy group;

R^4 represents a hydrogen atom or an amino-protecting group, such as a lower carboxylic acyl group;

R^5 represents a hydrogen atom or a lower carboxylic acyl group;

R^6 represents a hydrogen atom;

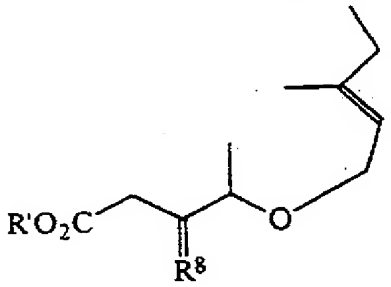
R^7 represents a hydrogen atom or, together with R, represents a lower alkylene group;

or

R, together with R^6, represents a lower alkylene group substituted with a hydroxy-loweralkenyl group;

or

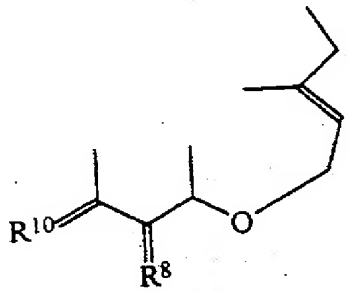
R, together with R^5 and R^6, represents a group of formula:



in which R^8 represents a hydroxyimino group or two hydrogen atoms, and R' represents a carboxy-protecting group;

or

R, together with R^4, R^5 and R^6, represents a group of formula (i):



(i)

in which R^8 is as defined above and R^{10} represents a keto group or two hydrogen atoms;

R^9 represents a hydroxy group, an amino group, an amino group substituted with one or two lower alkyl groups, an aryl-carboxylic acyl group optionally substituted with one or more substituents selected from Substituents (a) below, or an alkyl group, an alkenyl

group or alkynyl group which is straight or branched and which is optionally substituted with a substituent selected from:

sulfonyl groups, carboxyl groups, cyano groups, cycloalkyl groups, heteroaryl groups having from 5 to 10 ring atoms, 1 to 3 of which are selected from O, S and N, aryl and biaryl groups having from 1 to 13 carbon atoms and which may be further substituted with one or more substituents selected from Substituents (a) below;

n=0 or 1;

Substituents (a): halo, hydroxy, amino, nitro, azido and cyano groups;

or,

when the compound of formula (I) is a Wieland-Gumlich aldehyde (in which R^4 represents a hydrogen atom and R^5 represents a CHO group), a dimer thereof wherein the substituent R^4 from each monomer forms, together with the substituent R^5 from the other monomer, a methenylene link;

and pharmaceutically acceptable salts and esters thereof]

in therapy.

It will be appreciated that, where the compounds of formula (I) contain a quaternary amine group, then the compound may be balanced by any suitable cation. Non-exhaustive examples of such cations are: the halides; such as chloride, bromide and iodide; mesylate; and triflate, although other suitable cations will be readily apparent to those skilled in the art.

When the compound of formula (I) forms a Wieland-Gumlich aldehyde, then it is preferred that R^7 is a hydrogen atom. It is further preferred that the group represented by R^6 and R is an ethylene group and that the hydroxy-loweralkenyl group is 2-hydroxy-1-ethenyl spaced apart from R^5 by two carbon atoms.

Compounds of formula (I) wherein R^4, R^5, R^6 and R represent a group of the formula (i) are preferred, especially where R^8 represents two hydrogen atoms and R^{10} represents a keto group.

The dashed lines preferably both represent carbon-carbon bonds.

In compounds of formula (I), we prefer lower alkyl, lower alkenyl, lower alkoxy, lower alkylene and lower alkenylene to have from 1 to 4 carbon atoms, preferably from 1 to 3 carbon atoms. Particularly preferred alkyl groups are ethyl and methyl, especially methyl. The particularly preferred meaning for alkoxy is methoxy.

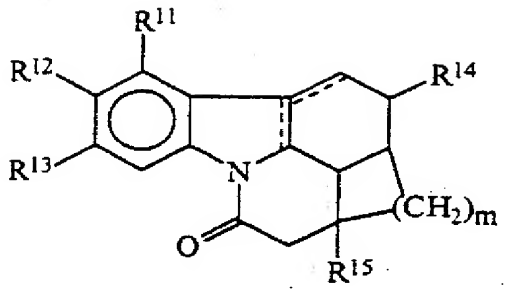
Where an alkyl, akenyl or alkynyl group is not specified as being "lower", then we prefer that it should have from 1 to 10 carbon atoms, preferably from 1 to 6 carbon atoms.

R^2 is preferably lower-alkoxy, hydrogen or hydroxy. When R^2 is lower-alkoxy, then we prefer that it should be methoxy.

While there are no especially preferred groups for R^1, the groups specified above for R^2 are particularly useful, especially where both R^1 and R^2 are the same and are both hydrogen or methoxy.

The present invention also provides the use of

compounds of formula (II):



(II)

[wherein the dashed lines optionally represent an additional carbon-carbon bond, provided that both do not represent additional carbon-carbon bonds;

R^{11}, R^{12} and R^{13} are the same or different and each represents a hydrogen atom, a hydroxy group, a carbamoyl group, a carbamoyl group substituted with one or two lower alkyl groups, a lower-alkoxy group, an aralkoxy group in which the aryl part has from 6 to 10 carbon atoms and the alkyl is a lower alkyl or an aryloxy group in which the aryl group has from 6 to 10 carbon atoms;

R^{14} represents a carboxy group, a carbamoyl group, a carbamoyl group substituted with one or two lower alkyl groups, or R^{14} represents a group of formula $-NR^{16}R^{17}$ wherein R^{16} and R^{17} are the same or different and each represents a hydrogen atom, a hydroxyalkyl group, a lower alkyl group optionally substituted by one or more substituents (a) as defined above, a carboxy-loweralkyl group, or R^{16} and R^{17} together represent an akylene group having from 3 to 6 carbon atoms and which may be substituted by substituents (a) as defined above, or one of R^{16} and R^{17} represents a hydrogen atom and the other represents an imino-loweralkyl group, a lower-alkoxy group, an aralkoxy group in which the aryl part has from

6 to 10 carbon atoms and the alkyl part is a lower alkyl, an aryloxy group in which the aryl group has from 6 to 10 carbon atoms, the above aryl parts being optionally substituted with one or more substituents selected from substituents (a) as defined above,

R^{15} represents a hydrogen atom, a lower alkyl group, an aryl group having from 6 to 10 carbon atoms, or an aralkyl group in which the aryl part has from 6 to 10 carbon atoms and the alkyl part is a lower alkyl;

and salts and esters thereof, including thioesters]

in therapy.

In the compounds of formula (II), m is preferably 2, 3 or 4, and is particularly preferably 3.

R^{15} is preferably a hydrogen atom.

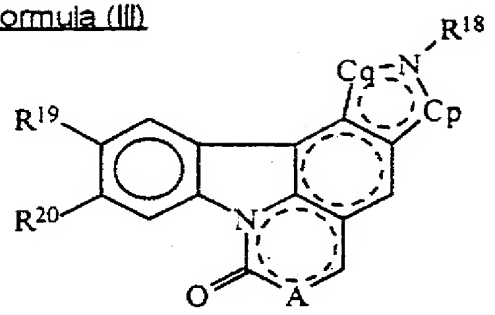
In general, we prefer that R^{11} represents either a hydrogen atom or a hydroxy group.

R^{12} and R^{13} preferably both represent hydrogen atoms. Alternatively, we prefer that one of R^{12} and R^{13} represents a hydroxy or a methoxy group and that the other represents a hydrogen atom or a hydroxy or methoxy group respectively.

The nature of the substituent at R^{14} is not important, provided that it is pharmaceutically acceptable and is suitable to derivatize the amino or carbonyl group, as appropriate.

The present invention also provides the use of compounds of formula (III):

Formula (III)



(III)

[wherein p+q=2, 3, or 4 or C_q-N-R^{18} is only attached to the remainder of the molecule by C_q and p=0;

rings containing dashed lines are saturated, partially unsaturated or completely unsaturated;

R^{18} represents a hydrogen atom, a lower-alkoxycarbonyl group or an aralkoxy group in which the aryl part has from 6 to 10 carbon atoms and the alkyl part is a lower alkyl and the aryl part is optionally substituted with one or more substituents selected from substituents (a) as defined above;

R^{19} and R^{20} are the same or different and each represents a hydrogen atom, a lower alkoxy group or a hydroxy group;

A represents =CH-, $-CH_2-$, -O- or -NH-;

and salts and esters thereof]

in therapy.

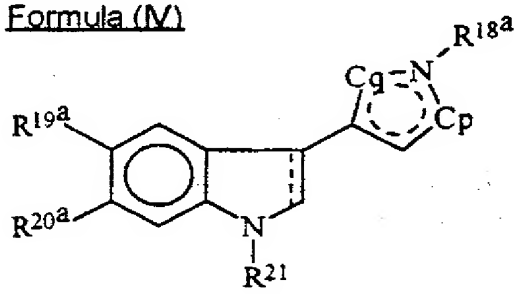
In the compounds of formula (III), we prefer that p+q=3.

The preferred meanings for R^{19} and R^{20} are as for R^1 and R^2 and R^{12} and R^{13} above.

In general, we prefer that A represents =CH- or $-CH_2-$.

The present invention further provides the use of compounds of formula (IV):

Formula (IV)



(IV)

[wherein the dashed ring indicates unsaturation, saturation, or partial unsaturation, and the dashed line represents an optional extra carbon-carbon bond;

R^{18a}, R^{19a} and R^{20a} have the same meanings as for R^{18}, R^{19} and R^{20} above, respectively;

R^{21} represents a hydrogen atom or a group of formula $-C(O)_r-A'-CH_2-Z$ in which r=0 or 1, A' is $-CH_2-$, -O- or -NH- and Z is a vinyl or ethynyl group;

and salts and esters thereof]

in therapy.

Preferred meanings for each of the substituents is generally as for the compounds of formula (III).

In the substituents R^{21}, we prefer that r=1.

The present invention also provides the use of compounds defined above in the manufacture of a medicament for the treatment of a condition responsive

to allosteric stimulation of muscarinic receptors.

It will be appreciated that the present invention also provides those compounds defined above which are novel.

The preferred muscarinic receptors are those classed as m1 - m5, especially m1 - m4, and those classed as M1 - M4. The most rpeferred receptor is the m1 receptor. Unless otherwise specified, these classifications are used interchangeably herein, except with regard to the central nervous system (CNS) wherein reference will normally be had to the m1 - m4 receptors.

Moreover, the preferred therapy is against dementia, particularly senile dementia and especially Alzheimer's Disease, which we believe to be particularly associated with the m1 receptor in the brain. Thus, we also prefer the condition responsive to allosteric stimulation of a muscarinic receptor to be Alzheimer's Disease.

Alternatively, we prefer to use the compounds of the present invention as sedatives for the CNS.

In the compounds of the present invention, where R^1, R^2, R^3, R^{16}, R^{17}, R^{19} or R^{20} represents a lower alkoxy group, this may be a straight or branched chain alkoxy group having from 1 to 4, preferably from 1 to 3, carbon atoms, and examples include the methoxy, ethoxy, propoxy, isopropoxy, butoxy, isobutoxy, sec-butoxy and t-butoxy groups, of which the most preferred are the methoxy and ethoxy groups.

Where R^1, R^2 or R^9 represents an amino group substituted with one or two lower alkyl groups, the alkyl groups may be straight or branched chain alkyl

groups having from 1 to 4, preferably from 1 to 3, carbon atoms, and examples include the methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl and t-butyl groups, of which the most preferred are the methyl and ethyl groups. Specific examples of such alkylamino groups include the methylamino, ethylamino, propylamino, isopropylamino, butylamino, isobutylamino, sec-butylamino, t-butylamino, dimethylamino, N-methyl-N-ethylamino, N-methyl-N-propylamino, N-methyl-N-isopropylamino, N-methyl-N-butylamino, N-methyl-N-isobutylamino, N-methyl-N-sec-butylamino, N-methyl-N-t-butylamino, N-ethyl-N-propylamino, N-ethyl-N-isopropylamino, N-ethyl-N-butylamino, N-ethyl-N-isobutylamino, N-ethyl-N-sec-butylamino, N-ethyl-N-t-butylamino, diethylamino, dipropylamino and dibutylamino groups.

Where R^1, R^2 or R^{14} represents a carbamoyl group substituted with one or two lower alkyl groups, the alkyl groups may be as defined and exemplified above in relation to the alkylamino groups. Specific examples of such alkylcarbamoyl groups include the methylcarbamoyl, ethylcarbamoyl, propylcarbamoyl, isopropylcarbamoyl, butylcarbamoyl, isobutylcarbamoyl, sec-butylcarbamoyl, t-butylcarbamoyl, dimethylcarbamoyl, N-methyl-N-ethylcarbamoyl, N-methyl-N-propylcarbamoyl, N-methyl-N-isopropylcarbamoyl, N-methyl-N-butylcarbamoyl, N-methyl-N-isobutylcarbamoyl, N-methyl-N-sec-butylcarbamoyl, N-methyl-N-t-butylcarbamoyl, N-ethyl-N-propylcarbamoyl, N-ethyl-N-isopropylcarbamoyl, N-ethyl-N-butylcarbamoyl, N-ethyl-N-isobutylcarbamoyl, N-ethyl-N-sec-butylcarbamoyl, N-ethyl-N-t-butylcarbamoyl, diethylcarbamoyl, dipropylcarbamoyl and dibutylcarbamoyl groups.

Where R^4 represents an amino-protecting group, there is no particular restriction on the nature of this

group and any such group commonly used for the protection of amino groups in this type of compound may equally be used here. Examples of suitable protecting groups include acyl groups, such as the lower aliphatic acyl or aromatic acyl groups, for example: aliphatic lower acyl groups such as the formyl, acetyl, propionyl, butyryl, isobutyryl, valeryl, isovaleryl and pivaloyl groups; and aromatic acyl groups, such as the benzoyl, 4-acetoxybenzoyl, 4-methoxybenzoyl, 4-methylbenzoyl and 1-naphthoyl groups. Of these, we prefer the acetyl, benzoyl and isobutyryl groups. Other protecting groups include alkyl groups, which may be straight or branched chain groups having from 1 to 6 carbon atoms, of which examples include the methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, t-butyl, pentyl, isopentyl, neopentyl, 2-methylbutyl, 1-ethylpropyl, 4-methylpentyl, 3-methylpentyl, 2-methylpentyl, 1-methylpentyl, 3,3-dimethylbutyl, 2,2-dimethylbutyl, 1,1-dimethylbutyl, 1,2-dimethylbutyl, 1,3-dimethylbutyl, 2,3-dimethylbutyl, 2-ethylbutyl, hexyl and isohexyl groups; of these, we prefer those alkyl groups having from 1 to 4 carbon atoms, preferably the methyl, ethyl, propyl, isopropyl, butyl and isobutyl groups, and most preferably the methyl group. Further amino-protecting groups include the aralkyl groups, in which the alkyl part has from 1 to 3 carbon atoms and the aryl part is a carbocyclic aromatic group having from 6 to 14, more preferably from 6 to 10, and most preferably 6 or 10, carbon atoms, which may be substituted or unsubstituted and, if substituted, has at least one of substituents α defined and exemplified above, although the unsubstituted groups are preferred; examples of such aralkyl groups include the benzyl, phenethyl, 1-phenylethyl, 3-phenylpropyl, 2-phenylpropyl, 1-naphthylmethyl, 2-naphthylmethyl, 2-(1-naphthyl)-ethyl, 2-(2-naphthyl)ethyl, benzhydryl (i.e. diphenylmethyl), triphenylmethyl, bis(o-nitro-

phenyl)methyl, 9-anthrylmethyl, 2,4,6-trimethylbenzyl, 4-bromobenzyl, 2-nitrobenzyl, 4-nitrobenzyl, 3-nitrobenzyl, 4-methoxybenzyl and piperonyl groups.

Substituents x include:

alkyl groups, such as those exemplified above, especially the methyl group;

alkoxy groups, such as those exemplified above, especially the methoxy group;

halogen atoms, such as the fluorine, chlorine, bromine and iodine atoms, especially the fluorine and chlorine atoms;

nitro, cyano, carboxyl, and hydroxy groups; and

alkylenedioxy groups having from 1 to 3 carbon atoms, such as the methylenedioxy, ethylenedioxy and propylenedioxy groups.

Where R^5 represents a lower carboxylic acyl group, this is preferably a lower aliphatic carboxylic acyl group, and more preferably an alkanoyl or alkenoyl group (most preferably alkanoyl), for example a formyl, acetyl, propionyl, butyryl, isobutyryl, valeryl, isovaleryl, pivaloyl, acryloyl, methacryloyl or propioloyl group, of which the acetyl, propionyl and pivaloyl groups are preferred.

Where R^7 and R together represent a lower alkylene group, this may be a straight or branched chain alkylene group having from 1 to 3 or from 1 to 4 carbon atoms. Examples of such groups include the methylene, ethylene, ethylidene, trimethylene, propylene, propylidene, isopropylidene, tetramethylene, butylidene, 1-methyl-

ethylene, 2-methylethylene, 1-methyltrimethylene, 2-methyltrimethylene and 3-methyltrimethylene groups, of which the methylene and ethylene groups are preferred.

Where R^6 and R together represent a lower alkylene group substituted with a lower hydroxyalkenyl group, the alkylene part may be any one of the alkylene groups defined and exemplified above. Examples of hydroxyalkenyl groups include the 1- and 2- hydroxyvinyl, 3-hydroxyallyl, 3-hydroxymethallyl, 1-, 2-, 3- and 4-hydroxybut-2-enyl, 1- and 2-, 3- and 4- hydroxybut-3-enyl groups, of which the 2-hydroxyvinyl and 3-hydroxymethallyl groups are preferred.

Where R' represents a protected carboxy group, there is no particular restriction on the nature of the carboxy-protecting group used, and any carboxy-protecting group known in the art may equally be used in this reaction. Non-limiting examples of such groups include:

alkyl groups having from 1 to 25 carbon atoms, more preferably from 1 to 6 carbon atoms, such as the methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, t-butyl, pentyl, isopentyl, neopentyl, 2-methylbutyl, 1-ethylpropyl, 4-methylpentyl, 3-methylpentyl, 2-methylpentyl, 1-methylpentyl, 3,3-dimethylbutyl, 2,2-dimethylbutyl, 1,1-dimethylbutyl, 1,2-dimethylbutyl, 1,3-dimethylbutyl, 2,3-dimethylbutyl, 2-ethylbutyl, hexyl, isohexyl, heptyl, octyl, nonyl, decyl, dodecyl, tridecyl, pentadecyl, octadecyl, nonadecyl, icosyl, henicosyl, docosyl, tricosyl, tetracosyl and pentacosyl groups, but most preferably the methyl, ethyl and t-butyl groups;

cycloalkyl groups having from 3 to 7 carbon atoms,

for example the cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl and cycloheptyl groups;

aralkyl groups, in which the alkyl part has from 1 to 3 carbon atoms and the aryl part is a carbocyclic aromatic group having from 6 to 14 carbon atoms, which may be substituted or unsubstituted and, if substituted, has at least one of substituents x defined and exemplified above, although the unsubstituted groups are preferred; examples of such aralkyl groups include the benzyl, phenethyl, 1-phenylethyl, 3-phenylpropyl, 2-phenylpropyl, 1-naphthylmethyl, 2-naphthylmethyl, 2-(1-naphthyl)-ethyl, 2-(2-naphthyl)ethyl, benzhydryl (i.e. diphenylmethyl), triphenylmethyl, bis(o-nitro-phenyl)methyl, 9-anthrylmethyl, 2,4,6-trimethyl-benzyl, 4-bromobenzyl, 2-nitrobenzyl, 4-nitrobenzyl, 3-nitrobenzyl, 4-methoxybenzyl and piperonyl groups;

alkenyl groups having from 2 to 6 carbon atoms, such as the the vinyl, allyl, 2-methylallyl, 1-propenyl, isopropenyl, 1-butenyl, 2-butenyl, 3-butenyl, 1-pentenyl, 2-pentenyl, 3-pentenyl, 4-pentenyl, 1-hexenyl, 2-hexenyl, 3-hexenyl, 4-hexenyl and 5-hexenyl groups, of which the vinyl, allyl, 2-methylallyl, 1-propenyl, isopropenyl and butenyl groups are preferred, the allyl and 2-methylallyl groups being most preferred.

halogenated alkyl groups having from 1 to 6, preferably from 1 to 4, carbon atoms, in which the alkyl part is as defined and exemplified in relation to the alkyl groups above, and the halogen atom is chlorine, fluorine, bromine or iodine, such as the 2,2,2-trichloroethyl, 2-haloethyl (e.g. 2-chloro-ethyl, 2-fluoroethyl, 2-bromoethyl or 2-iodoethyl), 2,2-dibromoethyl and 2,2,2-tribromoethyl groups;

substituted silylalkyl groups, in which the alkyl part is as defined and exemplified above, and the silyl group has up to 3 substituents selected from alkyl groups having from 1 to 6 carbon atoms and phenyl groups which are unsubstituted or have at least one substituent selected from substituents α defined and exemplified above, for example a 2-trimethylsilylethyl group;

phenyl groups, in which the phenyl group is unsubstituted or substituted, preferably with at least one alkyl group having from 1 to 4 carbon atoms or acylamino group, for example the phenyl, tolyl and benzamidophenyl groups;

phenacyl groups, which may be unsubstituted or have at least one of substituents α defined and exemplified above, for example the phenacyl group itself or the p-bromophenacyl group;

cyclic and acyclic terpenyl groups, for example the geranyl, neryl, linalyl, phytyl, menthyl (especially m- and p- menthyl), thujyl, caryl, pinanyl, bornyl, norcaryl, norpinanyl, norbornyl, menthenyl, camphenyl and norbornenyl groups;

alkoxymethyl groups, in which the alkoxy part has from 1 to 6, preferably from 1 to 4, carbon atoms and may itself be substituted by a single unsubstituted alkoxy group, such as the methoxymethyl, ethoxymethyl, propoxymethyl, isopropoxymethyl, butoxymethyl and methoxyethoxymethyl groups;

aliphatic acyloxyalkyl groups, in which the acyl group is preferably an alkanoyl group and is more preferably an alkanoyl group having from 2 to 6 carbon atoms, and the alkyl part has from 1 to 6,

and preferably from 1 to 4, carbon atoms such as the acetoxymethyl, propionyloxymethyl, butyryloxymethyl, isobutyryloxymethyl, pivaloyloxymethyl, 1-pivaloyloxyethyl, 1-acetoxyethyl, 1-isobutyryloxyethyl, 1-pivaloyloxypropyl, 2-methyl-1-pivaloyloxypropyl, 2-pivaloyloxypropyl, 1-isobutyryloxyethyl, 1-isobutyryloxypropyl, 1-acetoxypropyl, 1-acetoxy-2-methylpropyl, 1-propionyloxyethyl, 1-propionyloxypropyl, 2-acetoxypropyl and 1-butyryloxyethyl groups;

cycloalkyl-substituted aliphatic acyloxyalkyl groups, in which the acyl group is preferably an alkanoyl group and is more preferably an alkanoyl group having from 2 to 6 carbon atoms, the cycloalkyl substituent has from 3 to 7 carbon atoms, and the alkyl part has from 1 to 6, preferably from 1 to 4, carbon atoms, such as the (cyclohexylacetoxy)methyl, 1-(cyclohexylacetoxy)ethyl, 1-(cyclohexylacetoxy)propyl, 2-methyl-1-(cyclohexylacetoxy)propyl, (cyclopentylacetoxy)methyl, 1-(cyclopentylacetoxy)ethyl, 1-(cyclopentylacetoxy)propyl and 2-methyl-1-(cyclopentylacetoxy)propyl, groups;

alkoxycarbonyloxyalkyl groups, especially 1-(alkoxycarbonyloxy)ethyl groups, in which the alkoxy part has from 1 to 10, preferably from 1 to 6, and more preferably from 1 to 4, carbon atoms, and the alkyl part has from 1 to 6, preferably from 1 to 4, carbon atoms, such as the 1-methoxycarbonyloxyethyl, 1-ethoxycarbonyloxyethyl, 1-propoxycarbonyloxyethyl, 1-isopropoxycarbonyloxyethyl, 1-butoxycarbonyloxyethyl, 1-isobutoxycarbonyloxyethyl, 1-sec-butoxycarbonyloxyethyl, 1-t-butoxycarbonyloxyethyl, 1-(1-ethylpropoxycarbonyloxy)ethyl and 1-(1,1-dipropylbutoxycarbonyloxy)ethyl groups,

and other alkoxycarbonylalkyl groups, in which both the alkoxy and alkyl groups have from 1 to 6, preferably from 1 to 4, carbon atoms, such as the 2-methyl-1-(isopropoxycarbonyloxy)propyl, 2-(isopropoxycarbonyloxy)propyl, isopropoxycarbonyloxymethyl, t-butoxycarbonyloxymethyl, methoxycarbonyloxymethyl and ethoxycarbonyloxymethyl groups;

cycloalkylcarbonyloxyalkyl and cycloalkyloxycarbonyloxyalkyl groups, in which the cycloalkyl group has from 3 to 10, preferably from 3 to 7, carbon atoms, is mono- or poly- cyclic and is optionally substituted by at least one (and preferably only one) alkyl group having from 1 to 4 carbon atoms (e.g. selected from those alkyl groups exemplified above) and the alkyl part has from 1 to 6, more preferably from 1 to 4, carbon atoms (e.g. selected from those alkyl groups exemplified above) and is most preferably methyl, ethyl or propyl, for example the 1-methylcyclohexylcarbonyloxymethyl, 1-methylcyclohexyloxycarbonyloxymethyl, cyclopentyloxycarbonyloxymethyl, cyclopentylcarbonyloxymethyl, 1-cyclohexyloxycarbonyloxyethyl, 1-cyclohexylcarbonyloxyethyl, 1-cyclopentyloxycarbonyloxyethyl, 1-cyclopentylcarbonyloxyethyl, 1-cycloheptyloxycarbonyloxyethyl, 1-cycloheptylcarbonyloxyethyl, 1-methylcyclopentylcarbonyloxymethyl, 1-methylcyclopentyloxycarbonyloxymethyl, 2-methyl-1-(1-methylcyclohexylcarbonyloxy)propyl, 1-(1-methylcyclohexylcarbonyloxy)propyl, 2-(1-methylcyclohexylcarbonyloxy)propyl, 1-(cyclohexylcarbonyloxy)propyl, 2-(cyclohexylcarbonyloxy)propyl, 2-methyl-1-(1-methylcyclopentylcarbonyloxy)propyl, 1-(1-methylcyclopentylcarbonyloxy)propyl, 2-(1-methylcyclopentylcarbonyloxy)propyl, 1-(cyclopentylcarbonyloxy)propyl, 2-(cyclopentylcarbonyloxy)propyl, 1-(1-methylcyclopentylcarbonyloxy)ethyl,

1-(1-methylcyclopentylcarbonyloxy)propyl, adamantyloxycarbonyloxymethyl, adamantylcarbonyloxymethyl, 1-adamantyloxycarbonyloxyethyl and 1-adamantylcarbonyloxyethyl groups;

cycloalkylalkoxycarbonyloxyalkyl groups in which the alkoxy group has a single cycloalkyl substituent, the cycloalkyl substituent having from 3 to 10, preferably from 3 to 7, carbon atoms and mono- or poly- cyclic, for example the cyclopropylmethoxycarbonyloxymethyl, cyclobutylmethoxycarbonyloxymethyl, cyclopentylmethoxycarbonyloxymethyl, cyclohexylmethoxycarbonyloxymethyl, 1-(cyclopropylmethoxycarbonyloxy)ethyl, 1-(cyclobutylmethoxycarbonyloxy)ethyl, 1-(cyclopentylmethoxycarbonyloxy)ethyl and 1-(cyclohexylmethoxycarbonyloxy)ethyl groups;

terpenylcarbonyloxyalkyl and terpenyloxycarbonyloxyalkyl groups, in which the terpenyl group is as exemplified above, and is preferably a cyclic terpenyl group, for example the 1-(menthyloxycarbonyloxy)ethyl, 1-(menthylcarbonyloxy)ethyl, menthyloxycarbonyloxymethyl, menthylcarbonyloxymethyl, 1-(3-pinanyloxycarbonyloxy)ethyl, 1-(3-pinanylcarbonyloxy)ethyl, 3-pinanyloxycarbonyloxymethyl and 3-pinanylcarbonyloxymethyl groups;

5-alkyl or 5-phenyl [which may be substituted by at least one of substituents α, defined and exemplified above] (2-oxo-1,3-dioxolen-4-yl)alkyl groups in which each alkyl group (which may be the same or different) has from 1 to 6, preferably from 1 to 4, carbon atoms, for example the (5-methyl-2-oxo-1,3-dioxolen-4-yl)methyl, (5-phenyl-2-oxo-1,3-dioxolen-4-yl)methyl, (5-isopropyl-2-oxo-1,3-dioxolen-4-yl)methyl, (5-t-butyl-2-oxo-1,3-dioxolen-

4-yl)methyl and 1-(5-methyl-2-oxo-1,3-dioxolen-4-yl)ethyl groups; and

other groups, such as the phthalidyl, indanyl and 2-oxo-4,5,6,7-tetrahydro-1,3-benzodioxolen-4-yl groups.

Where R^9 represents an aryl-carboxylic acyl group, this may be any one of the aromatic acyl groups exemplifeid in relation to amino-protecting groups.

Where R^9 or R^{15} represents an alkyl group having from 1 to 6 carbon atoms, this may be a straight or branched chain group having from 1 to 6, preferably from 1 to 5, carbon atoms, and examples include the methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, t-butyl, pentyl, isopentyl, neopentyl, 2-methylbutyl, 1-ethylpropyl, 4-methylpentyl, 3-methylpentyl, 2-methylpentyl, 1-methylpentyl, 3,3-dimethylbutyl, 2,2-dimethylbutyl, 1,1-dimethylbutyl, 1,2-dimethylbutyl, 1,3-dimethylbutyl, 2,3-dimethylbutyl, 2-ethylbutyl, hexyl and isohexyl groups. Of these, we prefer those alkyl groups having from 1 to 4 carbon atoms, preferably the methyl, ethyl, propyl, isopropyl, butyl and isobutyl groups, and most preferably the methyl group.

Where R^9 represents an alkenyl group this is preferably a group having from 2 to 5 carbon atoms, this may be a straight or branched chain group having from 2 to 5, preferably 3 or 4, carbon atoms, and examples include the vinyl, allyl, methallyl, 1-propenyl, isopropenyl, 1-butenyl, 2-butenyl, 3-butenyl, 1-pentenyl, 2-pentenyl, 3-pentenyl and 4-pentenyl groups, of which the vinyl, allyl, methallyl, 1-propenyl, isopropenyl and butenyl groups are preferred, the allyl and 2-butenyl groups being most preferred.

Where R^9 represents an alkynyl group this is preferably a group having from 2 to 5 carbon atoms, this may be a straight or branched chain group having from 2 to 5, preferably 3 or 4, carbon atoms, and examples include the ethynyl, propargyl (2-propynyl), 1-propynyl, 1-butynyl, 2-butynyl, 3-butynyl, 1-pentynyl, 2-pentynyl, 3-pentynyl and 4-pentynyl groups, of which the propynyl and butynyl groups are preferred, the propargyl and 2-butynyl groups being most preferred.

Where a group represented by R^9 is substituted by a cycloalkyl group, this preferably has from 3 to 7 carbon atoms, and examples include the cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl and cycloheptyl groups.

Where a group represented by R^9 is substituted by a heteroaryl group, this is an aromatic heterocyclic group having, in one or more (preferably 1 or 2) rings from 5 to 10 ring atoms, of which from 1 to 3 are nitrogen and/or oxygen and/or sulphur atoms. The group may be a monocyclic group or it may consist of two or more rings attached to each other by fusion or by a spiro attachment. In the case of those groups having 3 hetero-atoms in a ring, we prefer that 1, 2 or 3 should be nitrogen atoms and, correspondingly, 2, 1 or 0 should be oxygen and/or sulfur atoms. In the case of those groups having 1 or 2 hetero-atoms in a ring, the hetero-atoms may be freely chosen from nitrogen, oxygen and sulfur atoms. Preferably, however, the group contains at least one nitrogen atom. The group may be substituted or unsubstituted and, if substituted, the substituents are selected from the group consisting of substituents α and oxygen atoms, as defined and exemplified above. This is preferably an aromatic heterocyclic group having from 5 to 8, more preferably 5 or 6, ring atoms, of which from 1 to 3 are hetero atoms,

said aromatic heterocyclic group being unsubstituted or being substituted by at least one of substituents α, preferably an alkyl group having from 1 to 6 carbon atoms (such as those exemplified above, preferably a methyl group); specific examples of such aromatic heterocyclic groups include the pyridyl, pyridazinyl, pyrazinyl, pyrrolyl, imidazolyl, triazolyl, tetrazolyl, quinolyl, isoquiolyl, phthalazinyl, quinoxalinyl, quinazolinyl, cinnolinyl, carbazolyl, isoindolyl, indolyl, indazolyl and purinyl groups.

Where a group represented by R^9 is substituted by an aryl group, this is a carbocyclic aromatic group having from 6 to 14, preferably from 6 to 10, more preferably 6 or 10, and most preferably 6, ring carbon atoms in one or more aromatic rings. The group may also be unsubstituted or substituted, and, if substituted, the substituents are selected from the group consisting of substituents α, defined and exemplified above. In the case of the substituted groups, there is no particular limitation on the number of substituents on the aryl group, except such as may be imposed by the number of substitutable positions or possibly by steric constraints, but, in general, from 1 to 5 substituents are preferred, from 1 to 4 being more preferred and 1, 2 or 3 being most preferred. Also, where the group is substituted, it is preferred that it should not be further substituted by a group which is also substituted by another aryl group. Specific examples of the unsubstituted aromatic groups include the phenyl, α- or β- naphthyl, indenyl, phenanthrenyl and anthracenyl groups, of which we prefer those aromatic hydrocarbon groups having from 6 to 10 ring carbon atoms, particularly the phenyl, α-naphthyl and β-naphthyl groups, the phenyl group being most preferred.

Where the aryl group is substituted, the

substituents are selected from the group consisting of substituents α and substituents (a), defined and exemplified above, and examples of substituted groups include the 2-methylphenyl, 3-methylphenyl, 4-methylphenyl, 2-methoxyphenyl, 3-methoxyphenyl, 4-methoxyphenyl, 2-nitrophenyl, 3-nitrophenyl, 4-nitrophenyl, 2-fluorophenyl, 3-fluorophenyl, 4-fluorophenyl, 2-chlorophenyl, 3-chlorophenyl and 4-chlorophenyl groups.

Where a group represented by R^9 is substituted by a biaryl group, each aryl part may be as defined and exemplified above, and specific preferred examples of such groups include the biphenylyl and binaphthylyl groups, which may be unsubstituted or substituted as defined above.

Where R^{11}, R^{12}, R^{13}, R^{16}, R^{17} or R^{18} represents an aralkoxy group, the alkoxy part has from 1 to 3 carbon atoms and the aryl part is a carbocyclic aromatic group having from 6 to 14, more preferably from 6 to 10, and most preferably 6 or 10, carbon atoms, which may be substituted or unsubstituted and, if substituted, has at least one of substituents α defined and exemplified above, although the unsubstituted groups are preferred; examples of such aralkoxy groups include the benzyloxy, phenethyloxy, 1-phenylethoxy, 3-phenylpropoxy, 2-phenylpropoxy, 1-naphthylmethoxy, 2-naphthylmethoxy, 2-(1-naphthyl)ethoxy, 2-(2-naphthyl)ethoxy, benzhydryloxy (i.e. diphenylmethoxy), triphenylmethoxy, bis(o-nitrophenyl)methoxy, 9-anthrylmethoxy, 2,4,6-trimethylbenzyloxy, 4-bromobenzyloxy, 2-nitrobenzyloxy, 4-nitrobenzyloxy, 3-nitrobenzyloxy, 4-methoxybenzyloxy and piperonyloxy groups.

Where R^{11}, R^{12}, R^{13}, R^{16} or R^{17} represents

an aryloxy group, this is a carbocyclic aromatic group having from 6 to 14, preferably from 6 to 10, more preferably 6 or 10, and most preferably 6, ring carbon atoms in one or more aromatic rings. The group may also be unsubstituted or substituted, and, if substituted, the substituents are selected from the group consisting of substituents α, defined and exemplified above. In the case of the substituted groups, there is no particular limitation on the number of substituents on the aryloxy group, except such as may be imposed by the number of substitutable positions or possibly by steric constraints, but, in general, from 1 to 5 substituents are preferred, from 1 to 4 being more preferred and 1, 2 or 3 being most preferred. Also, where the group is substituted, it is preferred that it should not be further substituted by a group which is also substituted by another aryl group. Specific examples of the unsubstituted aromatic groups include the phenoxy, α- or β- naphthyloxy, indenyloxy, phenanthrenyloxy and anthracenyloxy groups, of which we prefer those groups having from 6 to 10 ring carbon atoms, particularly the phenoxy, α-naphthyloxy and β-naphthyloxy groups, the phenoxy group being most preferred. Examples of substituted groups include the 2-methylphenoxy, 3-methylphenoxy, 4-methylphenoxy, 2-methoxyphenoxy, 3-methoxyphenoxy, 4-methoxyphenoxy, 2-nitrophenoxy, 3-nitrophenoxy, 4-nitrophenoxy, 2-fluorophenoxy, 3-fluorophenoxy, 4-fluorophenoxy, 2-chlorophenoxy, 3-chlorophenoxy and 4-chlorophenoxy groups.

Where R^{16} or R^{17} represents a hydroxyalkyl group, this preferably has from 1 to 6 carbon atoms, and may be a straight or branched chain group, preferably having from 1 to 4, carbon atoms. Examples include the hydroxymethyl, 1- or 2- hydroxyethyl, 1-, 2- or 3- hydroxypropyl, 1- or 2- hydroxy-2-methylethyl, 1-, 2-, 3- or 4- hydroxybutyl, 1-, 2-, 3-, 4- or 5- hydroxy-

pentyl or 1-, 2-, 3-, 4-, 5- or 6- hydroxyhexyl groups. Of these, we prefer those hydroxyalkyl groups having from 1 to 4 carbon atoms, preferably the hydroxymethyl, 2-hydroxyethyl, 3-hydroxypropyl and 4-hydroxybutyl groups, and most preferably the hydroxymethyl group.

Where R^{16} or R^{17} represents a carboxyalkyl group, this preferably has from 1 to 6 carbon atoms, and may be a straight or branched chain group, preferably having from 1 to 4, carbon atoms. Examples include the carboxymethyl, 1- or 2- carboxyethyl, 1-, 2- or 3-carboxypropyl, 1- or 2- carboxy-2-methylethyl, 1-, 2-, 3- or 4- carboxybutyl, 1-, 2-, 3-, 4- or 5- carboxy-pentyl or 1-, 2-, 3-, 4-, 5- or 6- carboxyhexyl groups. Of these, we prefer those carboxyalkyl groups having from 1 to 4 carbon atoms, preferably the carboxymethyl, 2-carboxyethyl, 3-carboxypropyl and 4-carboxybutyl groups, and most preferably the carboxymethyl group.

Where one of R^{16} and R^{17} represents an iminoalkyl group, this preferably has from 1 to 6 carbon atoms, and may be a straight or branched chain group, preferably having from 1 to 4, carbon atoms. Examples include the iminomethyl, 1- or 2- iminoethyl, 1-, 2- or 3- iminopropyl, 1- or 2- imino-2-methylethyl, 1-, 2-, 3- or 4- iminobutyl, 1-, 2-, 3-, 4- or 5- iminopentyl or 1-, 2-, 3-, 4-, 5- or 6- iminohexyl groups. Of these, we prefer those iminoalkyl groups having from 1 to 4 carbon atoms, preferably the iminomethyl, 2-iminoethyl, 3-iminopropyl and 4-iminobutyl groups, and most preferably the iminomethyl group.

Where R^{15} represents an aralkyl group, the alkyl part has from 1 to 3 carbon atoms and the aryl part is a carbocyclic aromatic group having from 6 to 14, more preferably from 6 to 10, and most preferably 6 or 10, carbon atoms, which may be substituted or unsubstituted

and, if substituted, has at least one of substituents x defined and exemplified above, although the unsubstituted groups are preferred; examples of such aralkyl groups include the benzyl, phenethyl, 1-phenylethyl, 3-phenylpropyl, 2-phenylpropyl, 1-naphthylmethyl, 2-naphthylmethyl, 2-(1-naphthyl)-ethyl, 2-(2-naphthyl)ethyl, benzhydryl (i.e. diphenylmethyl), triphenylmethyl, bis(o-nitro-phenyl)methyl, 9-anthrylmethyl, 2,4,6-trimethylbenzyl, 4-bromobenzyl, 2-nitrobenzyl, 4-nitrobenzyl, 3-nitrobenzyl, 4-methoxybenzyl and piperonyl groups.

Where R^{18} represents an alkoxycarbonyl group, this may be a straight or branched chain alkoxycarbonyl group having from 2 to 5, preferably from 2 to 4, carbon atoms, and examples include the methoxycarbonyl, ethoxycarbonyl, propoxycarbonyl, isopropoxycarbonyl, butoxycarbonyl, isobutoxycarbonyl, sec-butoxycarbonyl and t-butoxycarbonyl groups, of which the most preferred are the methoxycarbonyl and ethoxycarbonyl groups.

Without being restricted by theory, the present invention envisages the use of the compounds of the present invention in the treatment of the following conditions:

Alzheimer's Disease;
Parkinson's Disease;
Motion sickness;
Huntingdon's chorea;
Schizophrenia;
Depression;
Anxiety;
Sedation;
Analgesia;
Stroke;
Preanaesthetic;
Antispasmodic;

Irritable Bowel Syndrome;
Bladder - incontinence, retention;
Peptic ulcer disease;
Bronchitis/asthma/cronic obstructive airways disease;
Sinus bradycardia;
Pacemaker regulation;
Glaucoma;
Achalasia;
Symptomatic diffuse oesophageal spasm;
Biliary dyskinesia;
Scleroderma;
Diabetes mellitus;
Lower oesophageal incompetence;
Intestinal pseudo obstruction;
Regulation of sleep;
Control of pupil diameter; and
Non-ulcer dyspepsia.

The compounds of the present invention may be administered in any suitable fashion for the desired treatment. For example, the compounds of the present invention can be administered orally in the form of tablets, capsules, granules, powders or syrups, or parenterally by intravenous injection, or as suppositories or the like. These pharmaceutical formulations can be prepared by mixing the compounds of the present invention with one or more adjuvants, such as excipients (e.g. organic excipients including sugar derivatives, such as lactose, sucrose, glucose, mannitol or sorbitol; starch derivatives, such as corn starch, mashed potato, α-starch, dextrine or carboxymethyl starch; cellulose derivatives, such as crystalline cellulose, low hydroxypropyl-substituted cellulose, hydroxypropylmethyl cellulose, carboxymethyl cellulose, carboxymethyl cellulose calcium or internally bridged carboxymethyl cellulose sodium; gum arabic; dextran; and Pullulan; inorganic excipients including silicates, such

as light silicic acid anhydride, synthetic aluminium silicate or magnesium meta-silicic acid aluminate; phosphates, such as calcium phosphate; carbonates, such as calcium carbonate; and sulphates, such as calcium sulphate); lubricants (e.g. metal stearates, such as stearic acid, calcium stearate or magnesium stearate; talc; colloidal silica; waxes, such as beeswax or spermaceti; boric acid; adipic acid; sulphates, such as sodium sulphate; glycol; fumaric acid; sodium benzoate; DL-leucine; sodium salts of aliphatic acids; lauryl sulphates, such as sodium laurylsulphate or magnesium laurylsulphate; silicates, such as silicic acid anhydride or silicic acid hydrate; and the foregoing starch derivatives); binders (e.g. polyvinyl pyrrolidone, Macrogol; and similar compounds to the excipients described above); disintegrating agents (e.g. similar compounds to the excipients described above; and chemically modified starch-celluloses, such as Crosscarmelose sodium, sodium carboxymethyl starch or bridged polyvinyl pyrrolidone); stabilisers (e.g. p-hydroxybenzoates, such as methylparaben or propylparaben; alcohols, such as chlorobutanol, benzyl alcohol or phenylethyl alcohol; benzalkonium chloride; phenols, such as phenol or cresol; thimerosal; dehydroacetic acid; and sorbic acid); corrigents (e.g. sweeteners, vinegar or perfums, such as those conventionally used); diluents and the like.

The compounds of the present invention may also be administered by any other suitable route, such as: parenterally, intravenously, eye-drops, suppositories, dermal patch and sustained release formulations, using any suitable excipients, preservatives, flavourings, colourings and other ingredients as appropriate and/or desired.

The dose varies depending upon the condition and age

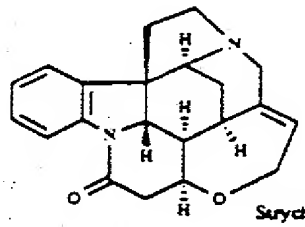
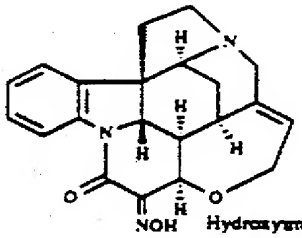
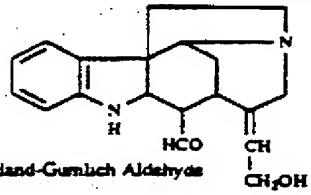
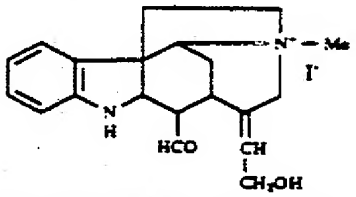
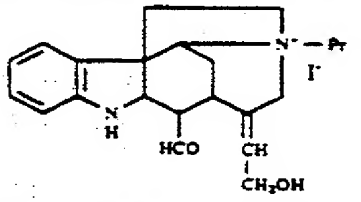
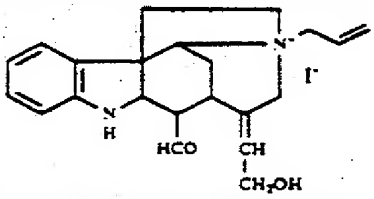
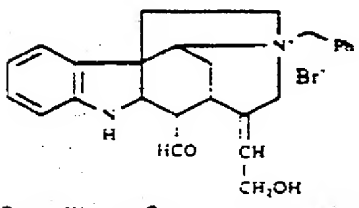
of the patient and upon the route and type of administration but, for example, the compounds of the present invention can be administered orally in a daily dose of from 0.01 to 1000 mg/kg body weight (preferably 0.05 to 200 mg/kg body weight), either as a single dose or as divided doses.

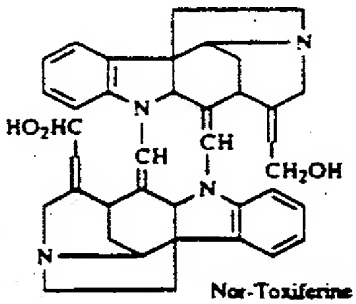
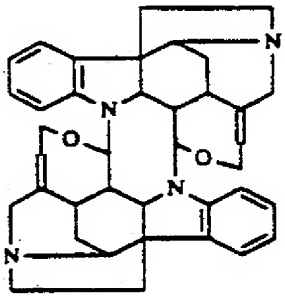
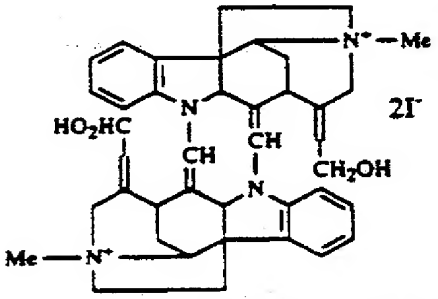
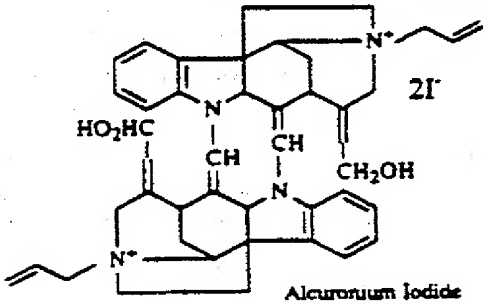
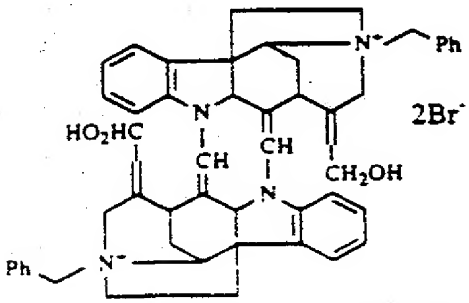
Various of the compounds of the present invention are shown in the following tables. In the tables, Bn represents a benzyl group.

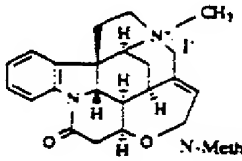
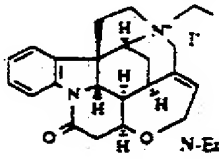
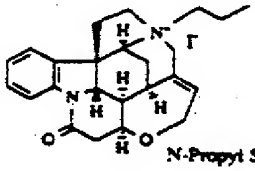
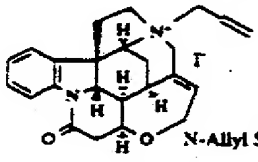
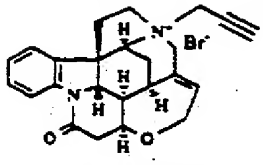
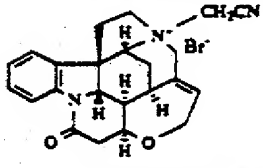
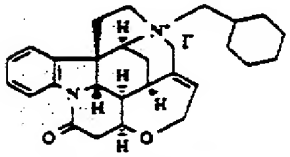
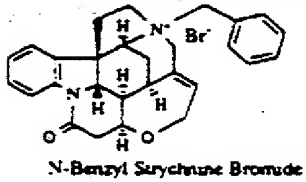
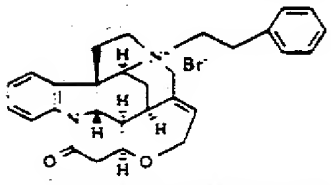
Following the tables are reaction schemes illustrating possible routes for the synthesis of various compounds of the present invention.

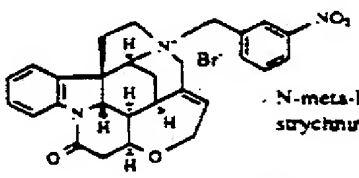
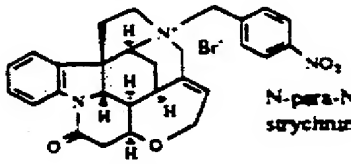
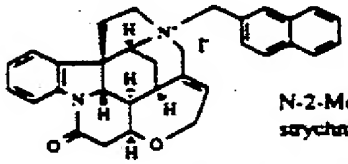
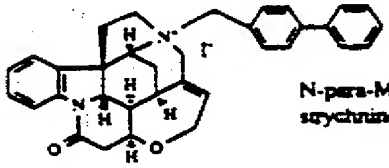
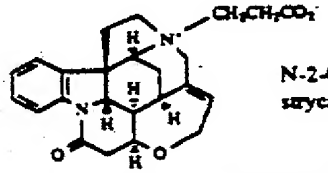
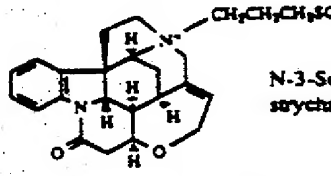
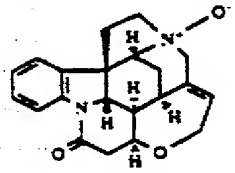
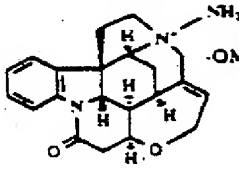
Compound numbers used in the reaction schemes correspond to those given in the preparative Examples following the reaction schemes and which illustrate the preparation of certain compounds of the invention.

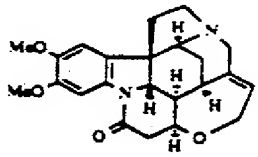
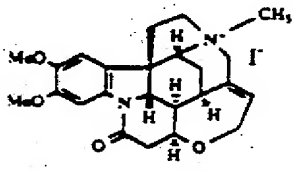
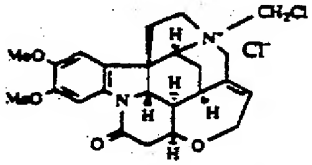
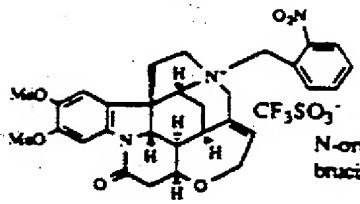
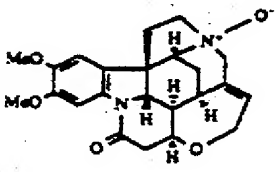
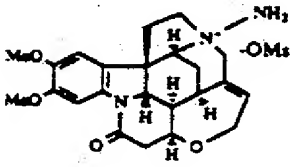
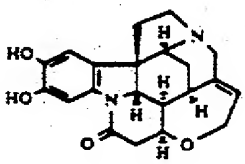
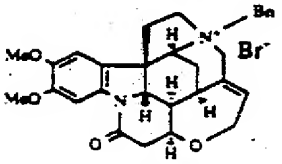
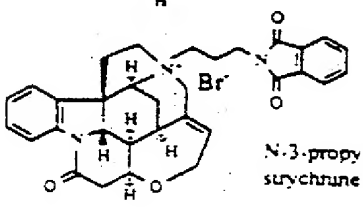
Finally, various methods for assaying the allosterism of the compounds of the invention are provided. The numbering in these methods and the tables which accompany them corresponds to the tables immediately following this text.

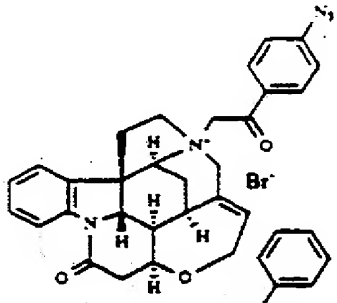
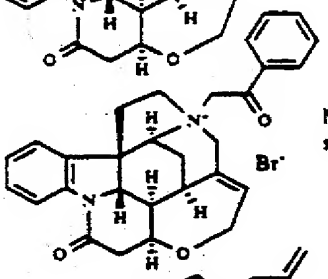
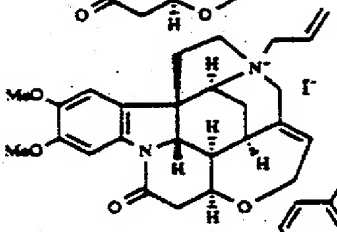
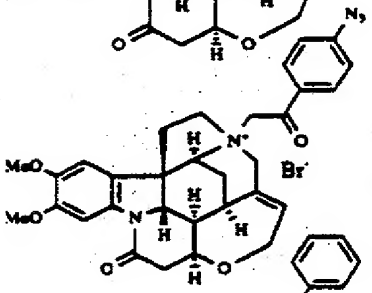
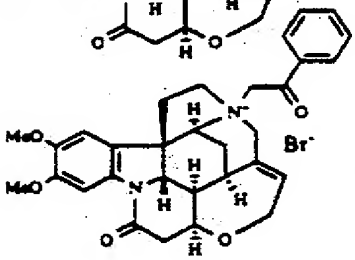
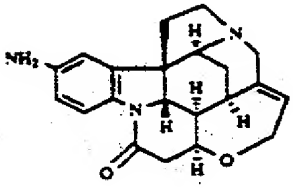
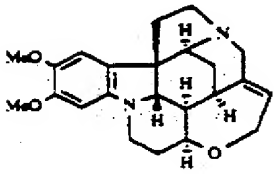
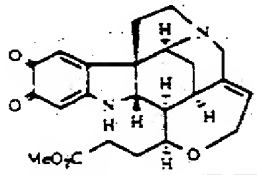
Compound no.	
001	 Strychnine
002	 Hydroxyimino strychnine
003	 Wieland-Gumlich Aldehyde
004	 N-Methyl Wieland-Gumlich Aldehyde iodide
005	 N-Propyl Wieland-Gumlich Aldehyde iodide
006	 N-Allyl Wieland-Gumlich Aldehyde iodide
007	 N-Benzyl Wieland-Gumlich Aldehyde bromide

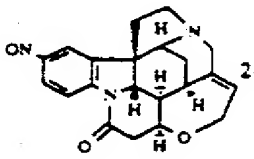
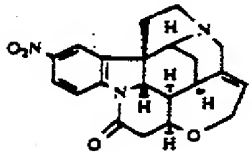
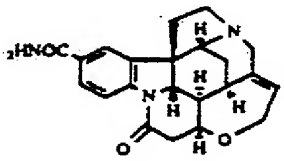
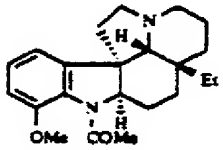
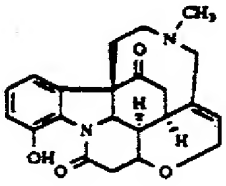
Compound no.	
008	 Nor-Toxiferine
009	
010	 N-Methyl Nor-Toxiferine Iodide
011	 Alcuronium Iodide
012	 N-Benzyl Nor-toxiferine Bromide

Compound no.	
013	 N-Methyl Strychnine Iodide
014	 N-Ethyl Strychnine Iodide
015	 N-Propyl Strychnine Iodide
016	 N-Allyl Strychnine Iodide
017	 N-Propargyl Strychnine Bromide
018	 N-Cyanomethyl Strychnine Bromide
019	 N-Cyclohexylmethyl Strychnine Iodide
020	 N-Benzyl Strychnine Bromide
021	 N-Phenylethyl Strychnine Bromide

Compound no.		
022		N-meta-Nitrobenzyl strychnine bromide
023		N-para-Nitrobenzyl strychnine bromide
024		N-2-Methylnaphthyl strychnine iodide
025		N-para-Methylbiphenyl strychnine iodide
026		N-2-Carboxyethyl strychnine
027		N-3-Sulphonylpropyl strychnine
028		Strychnine-N-oxide
029		N-Amino Strychnine mesylate

Compound no.		
033		Brucine
034		N-Methyl brucine iodide
035		N-Chloromethyl brucine chloride
036		N-ortho-Nitrobenzyl brucine triflate
037		Brucine-N-oxide
038		N-Amino brucine mesylate
039		2,3-Dihydroxy strychnine
040		N-Benzyl brucine bromide
055		N-3-propylphthalimido strychnine bromide

Compound no.		
056		N-para-Azido-phenacyl strychnine bromide
057		N-Phenacyl strychnine bromide
058		N-Allyl brucine iodide
059		N-para-Azido-phenacyl brucine bromide
060		N-Phenacyl brucine bromide
061		2-Amino strychnine
062		Brucinidine
063		

Compound no.	
064	 2-Nitroso strychnine
065	 2-Nitro strychnine
076	 2-Carboxamido strychnine
188	 Aspidospermine
214	 Vomicine
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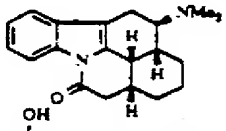
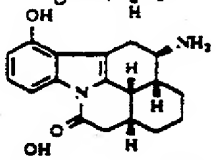
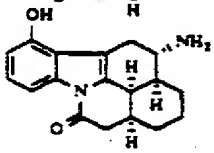
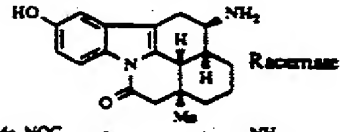
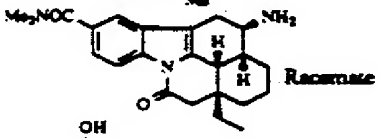
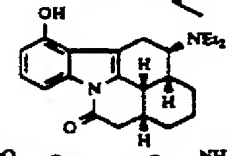
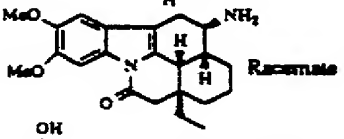
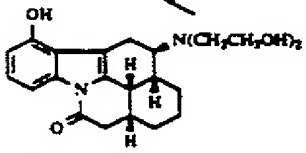
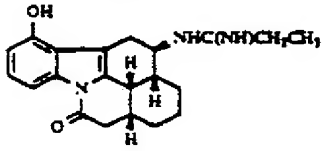
071	
072	
073	

Compound no.	
099	 Racemate
100	
101	
102	 Racemate
103	 Racemate
104	
105	 Racemate
106	
107	

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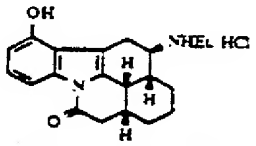
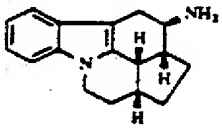
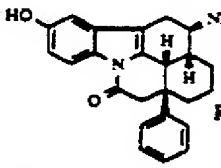
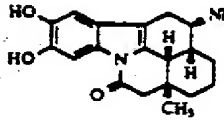
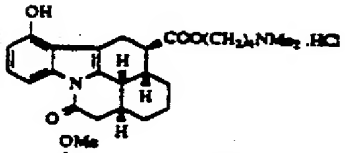
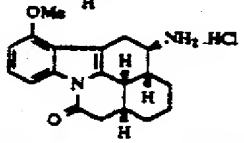
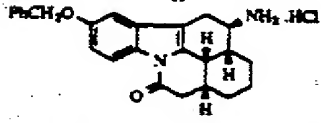
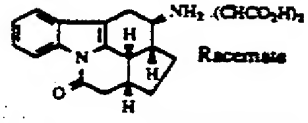
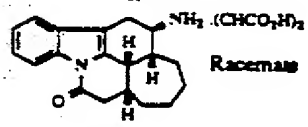
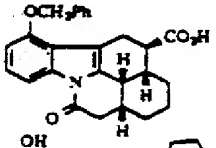
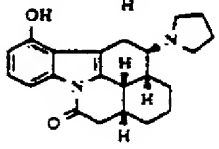
113	
114	 Racemate
115	 Racemate
116	 Racemate
118	
119	
120	
121	 Racemate
122	 Racemate
123	
124	

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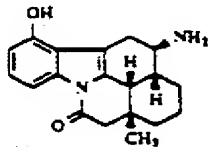
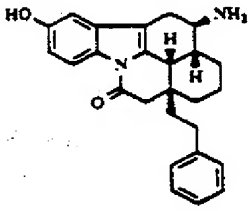
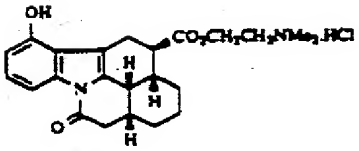
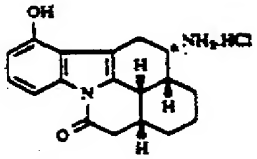
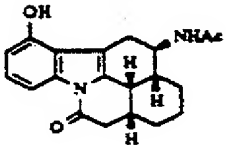
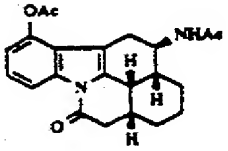
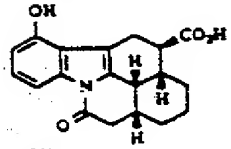
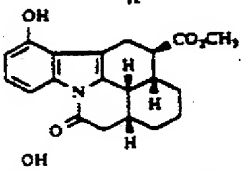
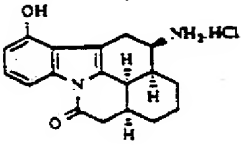
125	 Racemate
126	 Racemate
129	
130	
131	
132	
133	
134	
135	

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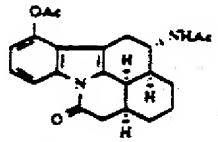
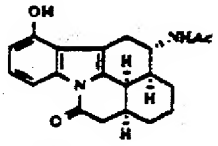
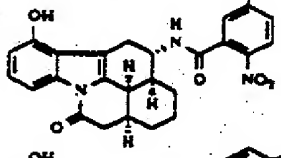
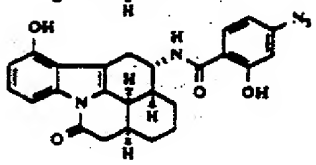
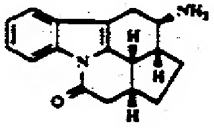
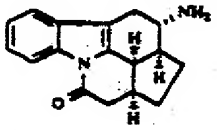
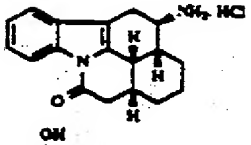
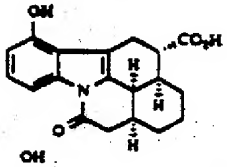
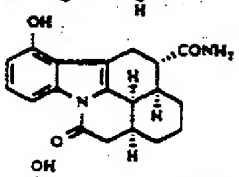
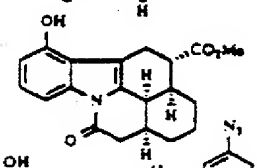
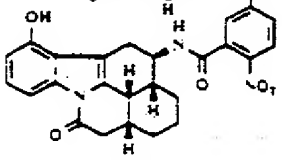
136	
137	
158	
159	
164	
165	
168	
169	
170	
171	
172	

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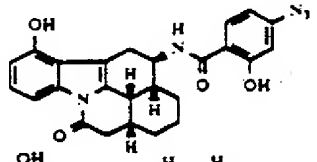
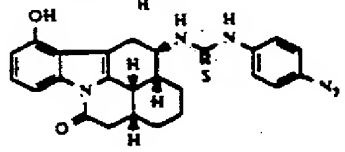
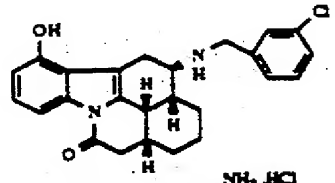
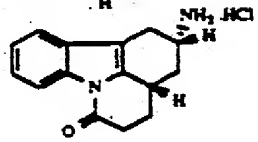
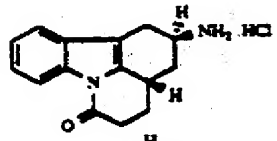
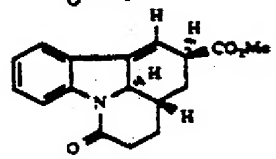
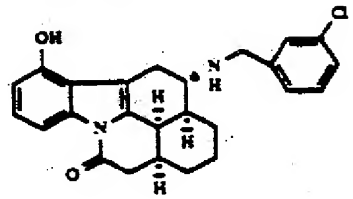
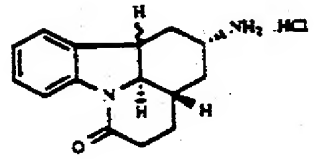
173	
174	
182	
183	
184	
185	
193	
201	

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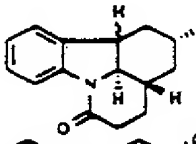
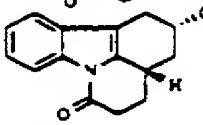
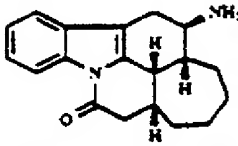
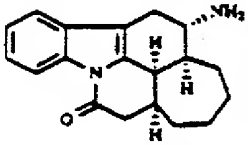
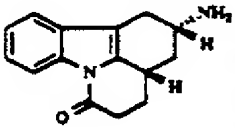
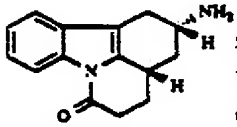
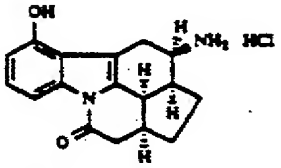
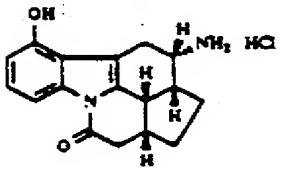
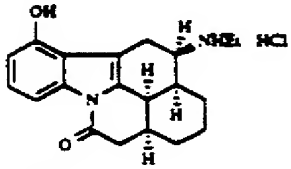
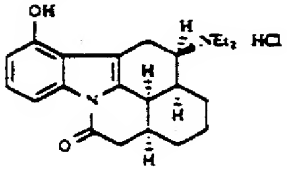
202	 Racemate
203	
204	 Single enantiomer Absolute stereoconfiguration unknown
205	 Single enantiomer Absolute stereoconfiguration unknown
206	 Single enantiomer Absolute stereoconfiguration unknown
207	 Single enantiomer Absolute stereoconfiguration unknown
246	
247	
248	
249	

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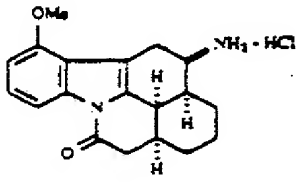
104	

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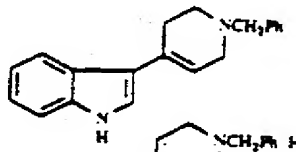
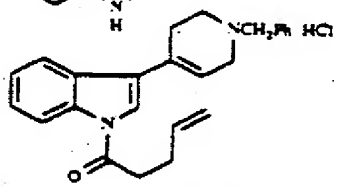
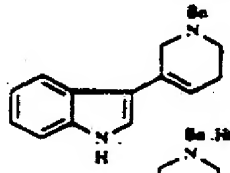
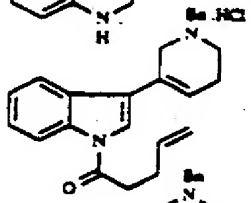
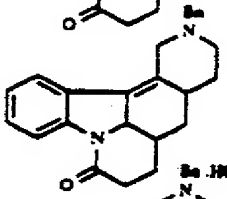
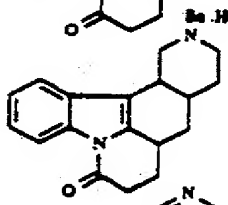
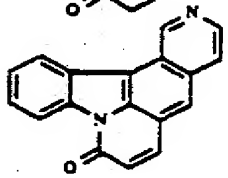
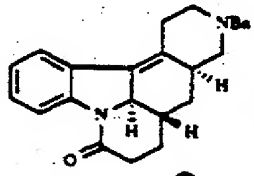
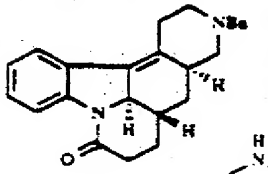
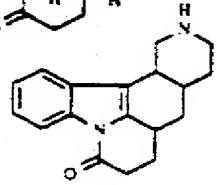
211	
212	
222	
223	
224	 Racemate
225	 Racemate
226	
227	 Single enantiomer Absolute stereoconfiguration unknown
228	 Single enantiomer Absolute stereoconfiguration unknown
229	 Racemate

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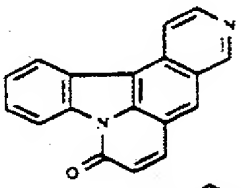
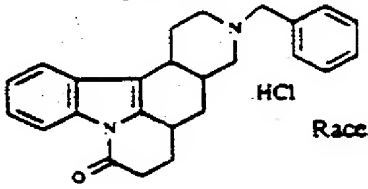
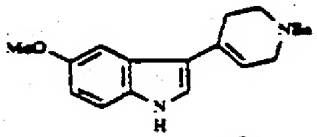
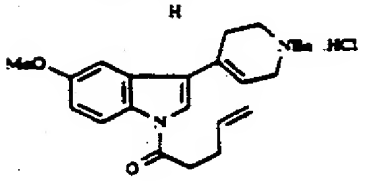
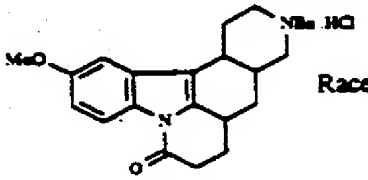
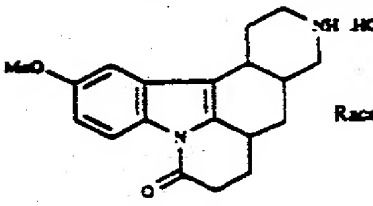
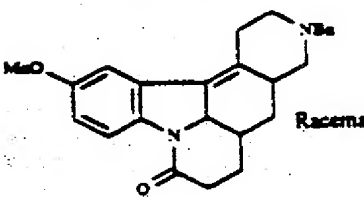
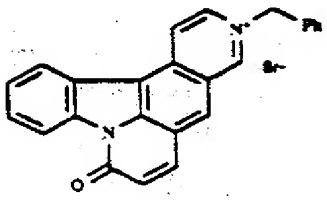
307	
308	 HCl Racemate
314	
315	
316	 Racemate
317	 Racemate
322	 Racemate
325	

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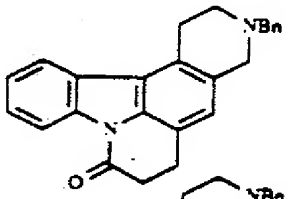
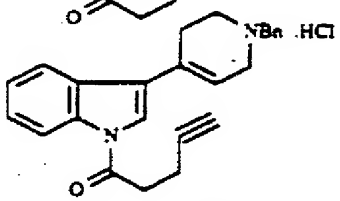
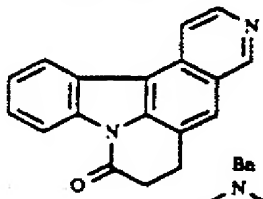
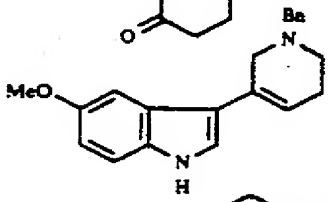
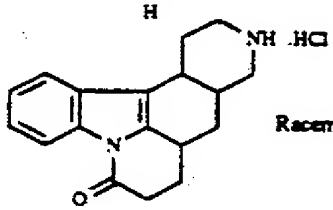
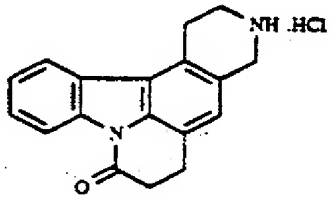
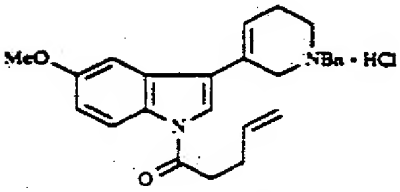
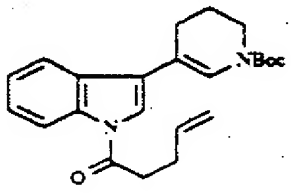
330	
331	
333	
334	
335	 Racemate
344	
381	
382	

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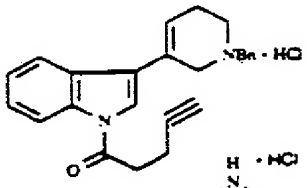
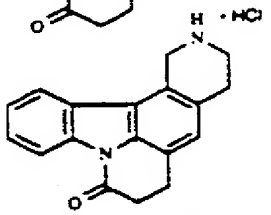
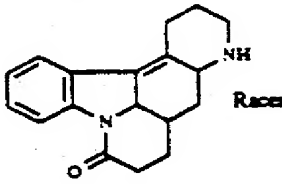
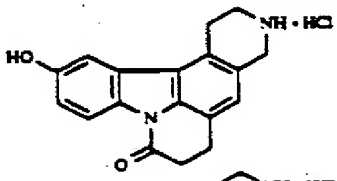
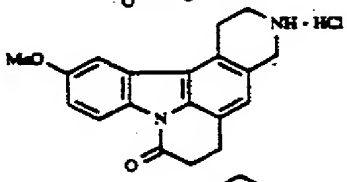
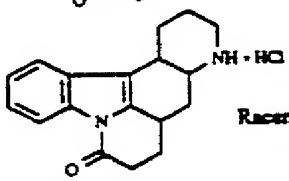
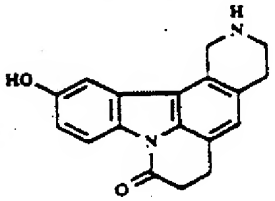
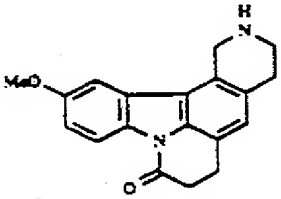
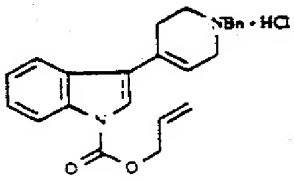
390	
391	
394	 Racemate
396	
397	
398	 Racemate
402	
403	
405	

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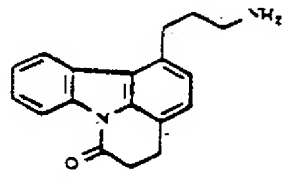
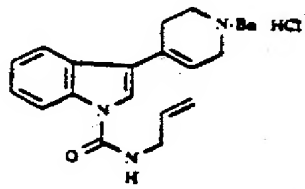
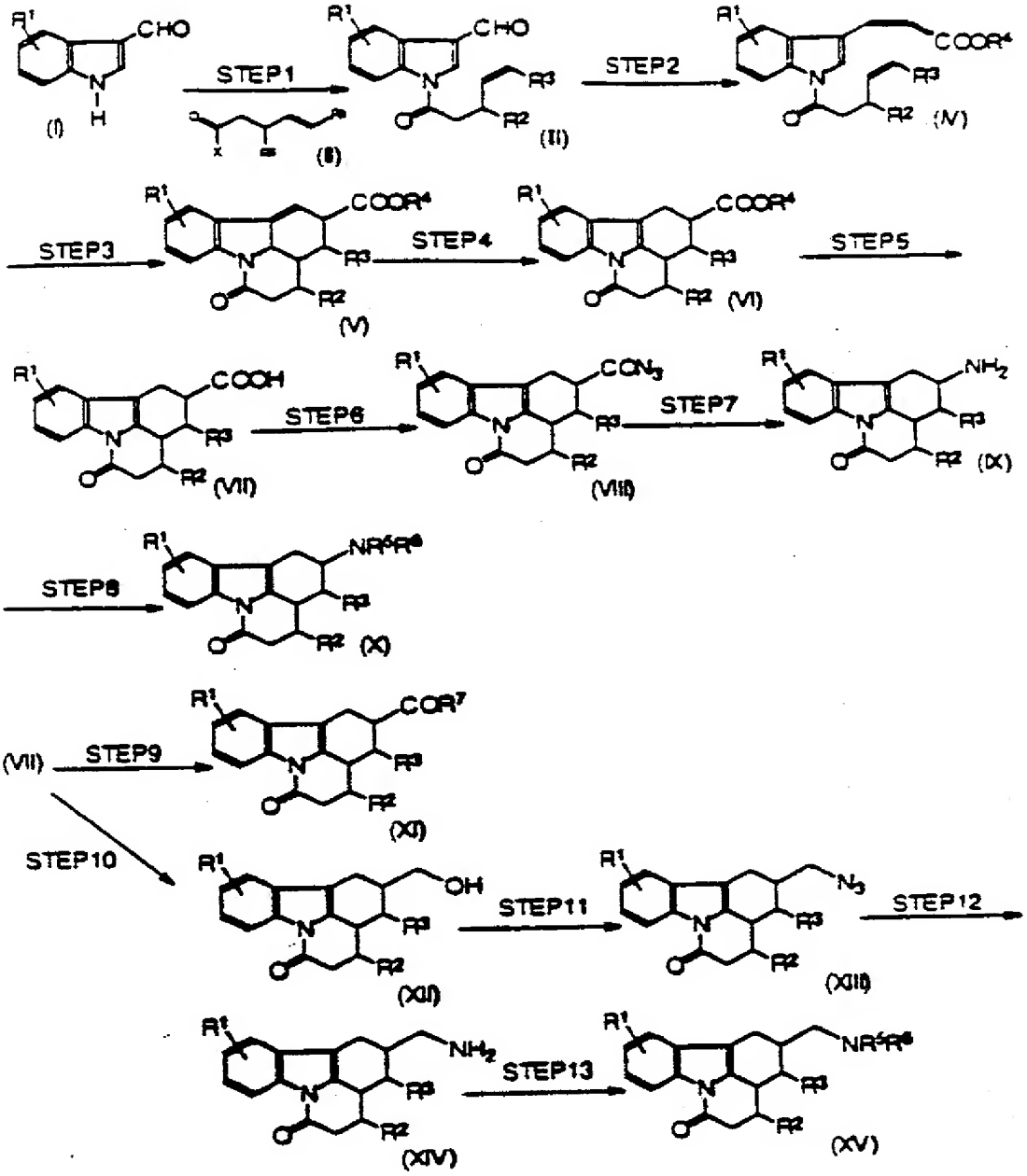
438	
439	



X represents halogen atoms or -OCOOR (preferably, Cl, Br, OCOOEt, OCOOiPr)

R^1 : hydroxy group, protected hydroxy group

R^2, R^3 each : hydrogen atom, alkyl group.

R^2 and R^3 together : $-(CH_2)_2-$, $-(CH_2)_3-$

R^4 : carboxy protecting group

R^5,R^6 : hydrogen atoms, amino protecting group
R^7 : NR^5R^6 -0-R^8,
where R^8 : unsubstituted alkyl group and alkyl group substituted with NR^5R^6

Step 1 :
(1) reagent
base : organic bases such as Et_3N or strong inorganic bases such as NaH, KN (trimethylsilyl)$_2$ (preferably, inorganic bases)
(2) solvent : ethers or DMF (preferably, dimethoxyethane, THF)
(3) temperature : -100 - room temperature (preferably, -70 -20)

Step 2 : Horner-Emmons reaction or Wittig reaction. Normally, trans compound is synthesized according to the above reaction. If you need cis compound, you have to follow the modified Horner-Emmons reaction
(W.Clark Still and Cesare Gennari, Tetrahedron Letters, Vol.24, no. 41, pp4405-44087 (1983)).
(1) reagent :
(a) ester : phosphonates such as $(R'O)_2POCH_2COOR$ or phosphonium salts such as $(Ph)_3$-$P^+(X^-)CH_2COOR$ (preferably, when synthesizing cis acid compound, $(CF_3CH_2O)^2$-PO-CH_2COOCH_3; when synthesizing trans acid compound, $(CH_3O)_2$-PO-CH_2COOCH_3
(b) base : NaH, KN(trimethylsilyl)$_2$. BuLi etc. (preferably, when synthesizing cis acid compound, KN(trimethylsilyl)$_2$ and 18-crown-6 ether; when synthesizing trans acid compound, NaH)
(2) solvent : ethers, DMF, aromatic hydrocarbons such as benzene (preferably, THF)
(3) temperature : -100 - 50 (preferably, when synthesizing cis acid compound, -70 -20; when synthesizing trans acid compound, 0 - r.t)

Step 3 :
(1) reagent : nothing
(a) solvent : aromatic hydrocarbons such as benzene, ethers (preferably, diphenylether, mesithylene, dichlorobenzene)
(3) temperature : 100 - 300 (preferably, 150 - 250) in the case of cis compound, 200 - 270 is preferable

Step 4:
(1) reagent :
(a) in case of using acid : preferably, mineral acids such as hydrochloric acid
(b) in the case of using metal : Pd, Rh (preferably, Pd-C)
(2) solvent : alcohols, aromatic hydrocarbons (preferably, ethanol)
(3) temperature : r.t. - 150 (preferably, 50 - 100)

Step 5:
(A) in the case of R^4=alkyl such as Me, Et; normal hydrolysis with alkaline
(1) reagent : metal hydroxide (preferably, NaOH, KOH)
(2) solvent : alcohols, water, water plus water soluble organic solvents
(3) temperature : 0 - 100 (preferably, r.t. - 80)

(B) in the case of R^4=benzyl derivatives; normal hydrogenolysis
(1) reagent : metal catalysts such as Pd-C, PtO_2 (preferably Pd-C)
(2) solvent: ethers such as THF or alcohols (preferably, ethanol, methanol)
(3) temperature : 0- 50 (preferably, r.t.)

(C) in the case of R^4=allyl
(1) reagent : metal catalyst (preferably, Pd catalyst such as $Pd(PPh_3)_4$)
(2) solvent : esters (preferably, ethyl acetate)
(3) temperature : 0 - 100 (preferably, r.t. - 50)

Step 6 : this step is comprised of a mixed anhydride method or an acyl halide method, followed by azidation.
(A) in the case of a mixed anhydride method :
(1) reagent : alkyl haloformates (XCOOR) (preferably, alkyl chloroformate; more preferably, ethyl chloroformate or i-butyl chloroformate), and organic amines (preferably, triethylamine)

(B) in the case of an acyl halide method :
(1) reagent : halogenation agents such as $SOCl_2$, PCl_5 (preferably, $SOCl_2$)

(C) azidation :
(1) reagent : alkali metal azide, phosphoryl azide (preferably, sodium azide)

(2) solvent : a mixture of water and organic solvent selected from ketones, ethers and alcohols (preferably, a mixture of water and organic solvent selected from ketones; more preferably, a mixture of water and acetone)

(3) temperature : -10 - 50 (preferably, 0 - r.t.)

Step 7 : This step is for preparing an amide compound and also for optical resolution, if necessary, and contains three reactions:

(A) Curtius Rearrangement according to the procedures described in P.A.S.Smith, Organic Reaction III, 337 (1946).
(B) Isocyanate to Carbamate by reacting methylbenzyl alcohol
(C) if necessary, optical resolution
(D) Elimination of protecting groups (Carbamate to Amine)

(A) and (B)

(1) reagent : arakyl alcohols (preferably, benzyl alcohol, 1-phenethylalcohol, p-methoxybenzylalcohol)
(2) solvent : aromatic hydrocarbons, amides, ethers (preferably, toluene)
(3) temperature : 50 - 200 (preferably, 70 150)

(C) normal optical resolution

(D-1) when the protecting group is a benzyl group : hydrogenolysis

(1) reagent : metal catalyst such as Pd-C, PtO_2 (preferably, Pd-C)
(2) solvent : alcohols, ethers, amides such as DMF (preferably, alcohols, amides; more preferably, ethanol, DMF)
(3) temperature : 0 - 50 (preferably, r.t.)

(D-2) when the protecting group is an alkyl group :

(1) reagent : BBr_3 or HI (preferably, BBr_3)
(2) solvent : halogenated hydrocarbons (preferably, CH_2Cl_2)
(3) temperature : -20 - 80 (preferably, 0 - 50)

Step 8 : acylation or alkylation

(1) reagent : acylating agents or alkylating agents, and bases
acylating agents or alkylating agents : alkyl halides or alkyl sulfonates (preferably, alkyl iodide)
bases : alkali metal salts, organic amines (preferably, $NaHCO_3$, K_2CO_3)
(2) solvent : aromatic hydrocarbons, alcohols (preferably, aromatic hydrocarbons; more preferably, toluene, benzene)
(3) temperature : r.t. - 200 (preferably, 50 - 150)

Step 9 :

(A) Via acyl halide (amidation and esterification)

(1) reagent :
(a) halogenating agents such as $SOCl_2$, PCl_5, $POCl_3$ (preferably, $SOCl_2$)
(b) bases : organic amines (preferably, pyridine, triethylamine)
(c) HNR^5R^6 or $HO\text{-}R^8$

(2) solvent : no solvent, or halogenated hydrocarbons or aromatic hydrocarbons (preferably, alcohols halogenated hydrocarbons; more preferably, CH_2Cl_2, $CHCl_3$)

(3) temperature : 0 - 80 (preferably, 20 - 50)

(B) Via acid anhydride (amidation and esterification)

(1) reagent :
(a) alkyl haloformates (XCOOR) (preferably, alkyl chloroformate; more preferably, ethyl chloroformate or 1-butyl chloroformate)
(b) bases : organic amines (preferably, pyridine, triethylamine)
(c) HNR^5R^6 or $HO\text{-}R^8$

(2) solvent : halogentated hydrocarbons, ketones or ethers (preferably, ketones and ethers; more preferably, acetone, THF)

(3) temperature : -20 - 80 (preferably, 0 - 50)

(C) Via direct esterification using diazomethane

(1) reagent : diazomethane

(2) solvent : halogenated hydrocarbons, ketones, alcohols or ethers (preferably, ketones and alcohols; more preferably, acetone, methanol)

(3) temperature : -20 - 80 (preferably, 0 - 50)

Step 10 : mild reduction

(1) reagent : BH_3-THF complex or other BH_3 complex (preferably, BH_3-THF complex)
(2) solvent : ethers (preferably, THF)
(3) temperature : -20 - 50 (preferably, 0 - r.t.)

Step 11 : This step is comprised of two reactions, that is, sulfonylation and azidation

(A) Sulfonylation
(1) reagent : sulfonyl halides such as sulfonyl chlorides preferably, CH_3SO_2Cl, CH_3-Ph-SO_2Cl) and bases such as organic amines (preferably, pyridine, triethylamine)
(2) solvent : pyridine, ethers, halogenated hydrocarbons (preferably, pyridine)
(3) temperature : -20 - 50 (preferably, 0 - r.t.)

(B) azidation
(1) reagent : TMS-N_3+$Bu_4N^+F^-$), metal azide such as NaN_3 (preferably, TMS-N_3+$Bu_4N^+F^-$)
(2) solvent : ethers, amides such as DMF and DMSO (preferably, ethers; more preferably, THF)
(3) temperature : r.t. - 200 (preferably, 50 - 150)

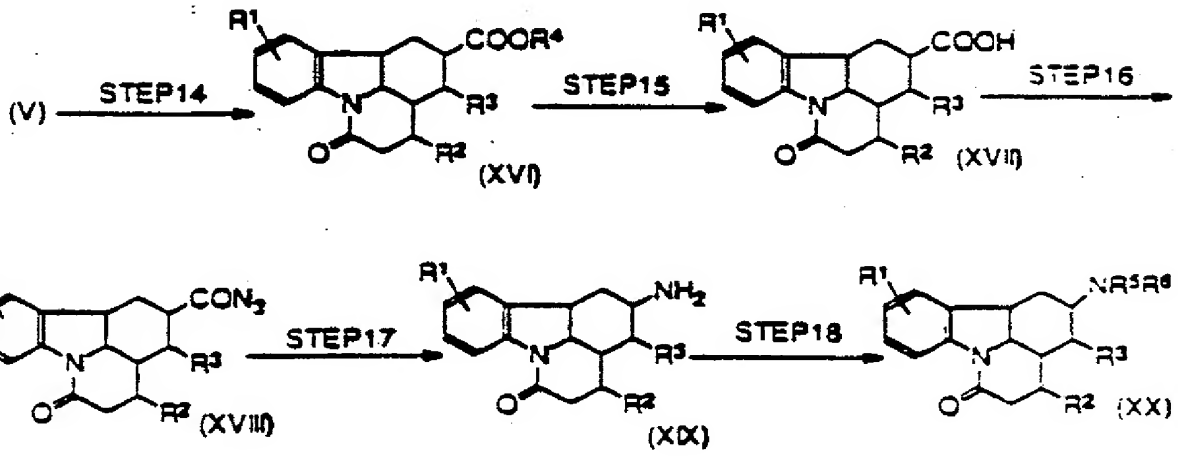
Step 12 : reduction

(1) reagents :
(a) metal catalyst such as Pd-C, PtO_2
(b) mineral acids such as hydrochloric acid

(2) solvent : alcohols, ethers, amides (preferably, alcohols; more preferably, ethanol)

(3) temperature : 0 - 50 (preferably, r.t.)

Step 13 : same as Step 8



Step 14 : reduction
(1) reagents : metal catalyst such as Pd-C, PtO_2
(2) solvent : alcohols, ethers, amides (preferably, alcohols; more preferably, ethanol)
(3) temperature : 0 - 50 (preferably, r.t.)

Step 15 :
(1) reagent : metal hydroxide (preferably, NaOH, KOH)
(2) solvent : alcohols, water, water plus soluble organic solvents
(3) temperature : 0 - 100 (preferably, r.t. - 80)

Step 16 : same as Step 6

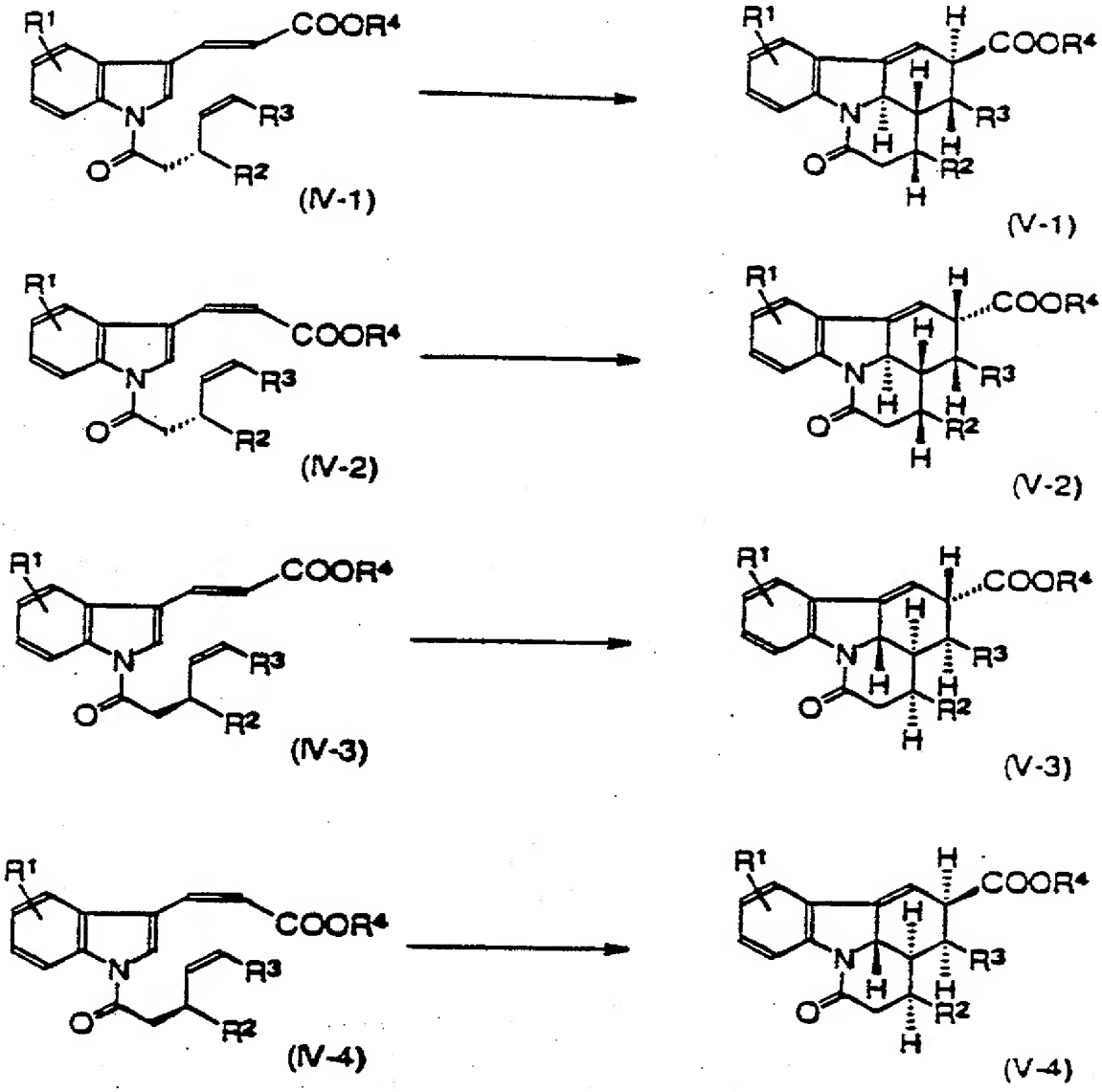
Step 17 : same as Step 7

Step 18 : same as Step 8

Optionally, the hydroxy protecting group of R^1 can be eliminated in Steps 4, 5, 8, 9, 10, 12, 13, 14, 15, 17 and 18.

d,l isomers and optical isomers of compound(II) can be obtained according to the procedures described in the following papers :
(1) T.Fukazawa, T.Hashimoto, Tetrahedron Asymmetry,4,11, 2323-2326(1993).
(2) JP Kokai hei 3-163078
(3) JP Kokai sho 61-27986
(4) JP Kokai sho 61-282384
(5) JP Kokai hei 4-182453
(6) JP Kokai sho 62-81383
(7) Y.Shimoji,K.Tomita,T.Hashimoto,F.Saito,Y.Morisawa, H.Mizuno,R.Yorikane, and H.Koike, J.Med.Chem.,35,816-822(1992).

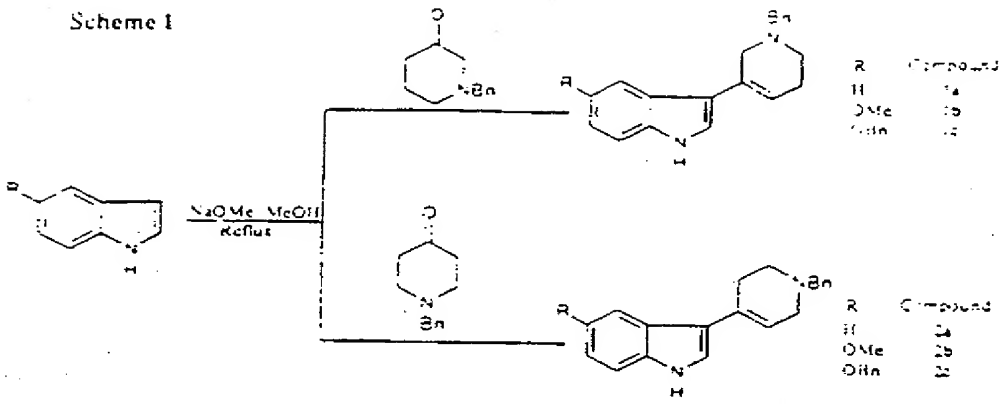
Stereochemistry of the present compounds



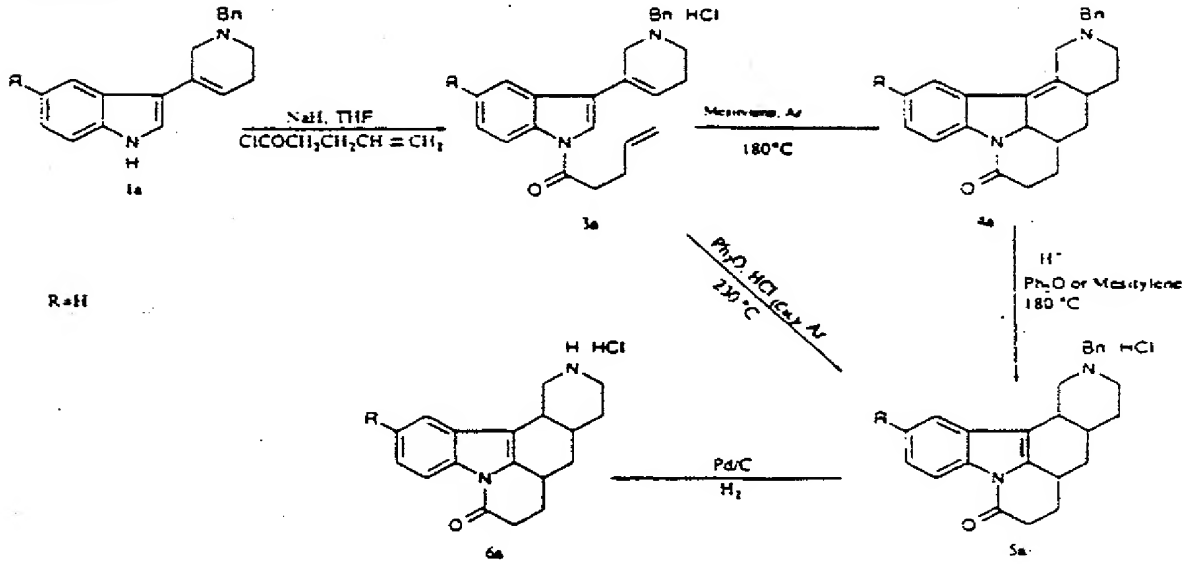
Enantiomers : (IV-1)&(IV-3), (IV-2)&(IV-4), (V-1)&(V-3), (V-2)&(V-4)

Epimers : (V-1)&(V-2), (V-3)&(V-4)

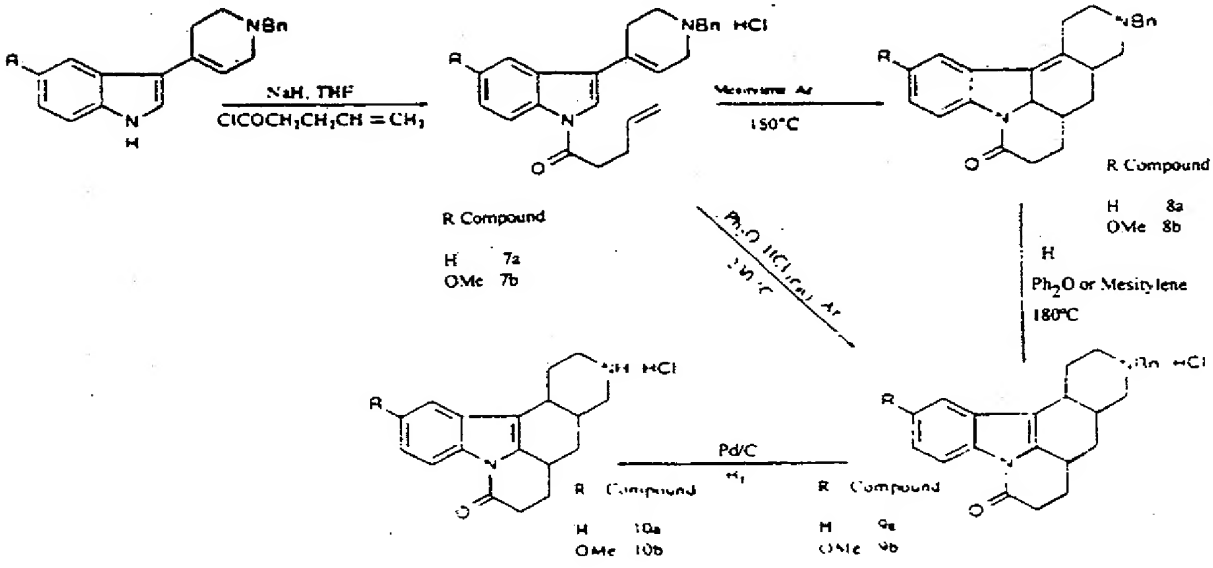
Scheme 1



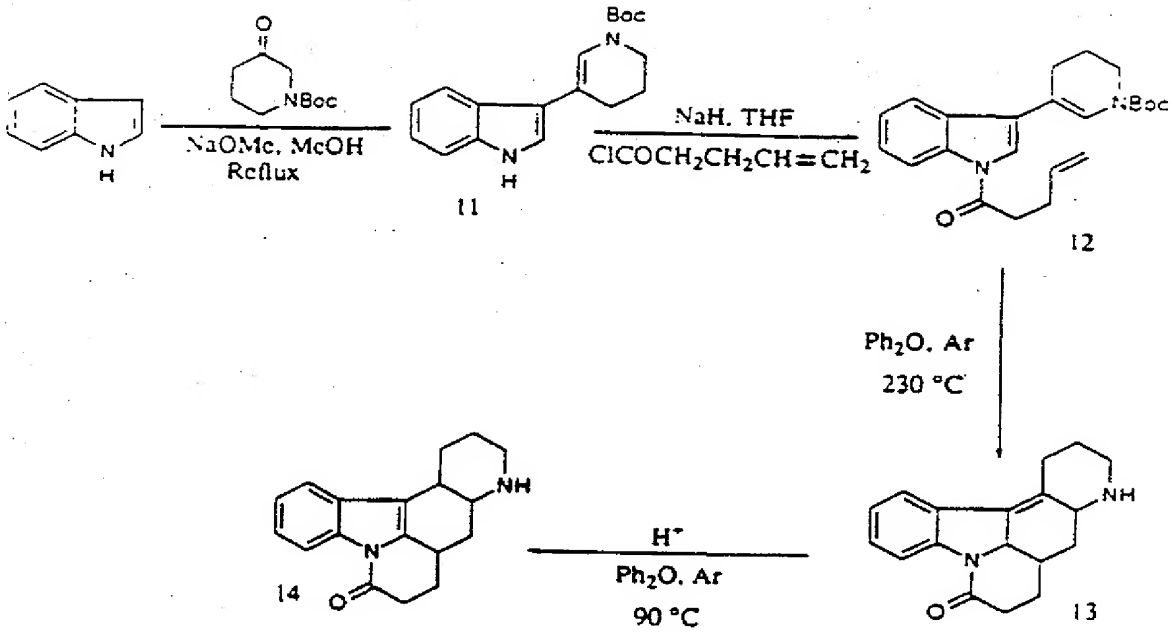
Scheme 2



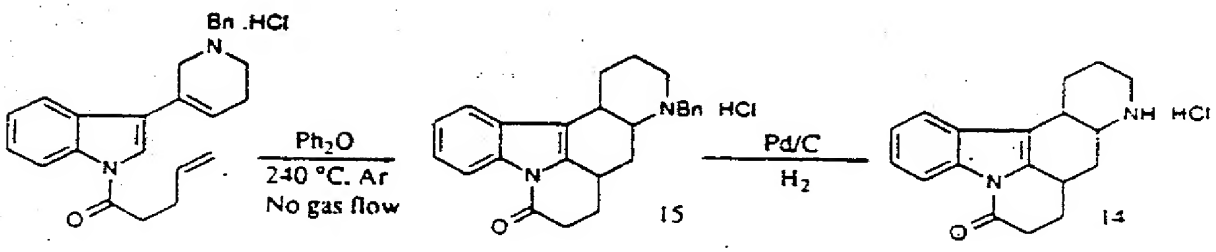
Scheme 3



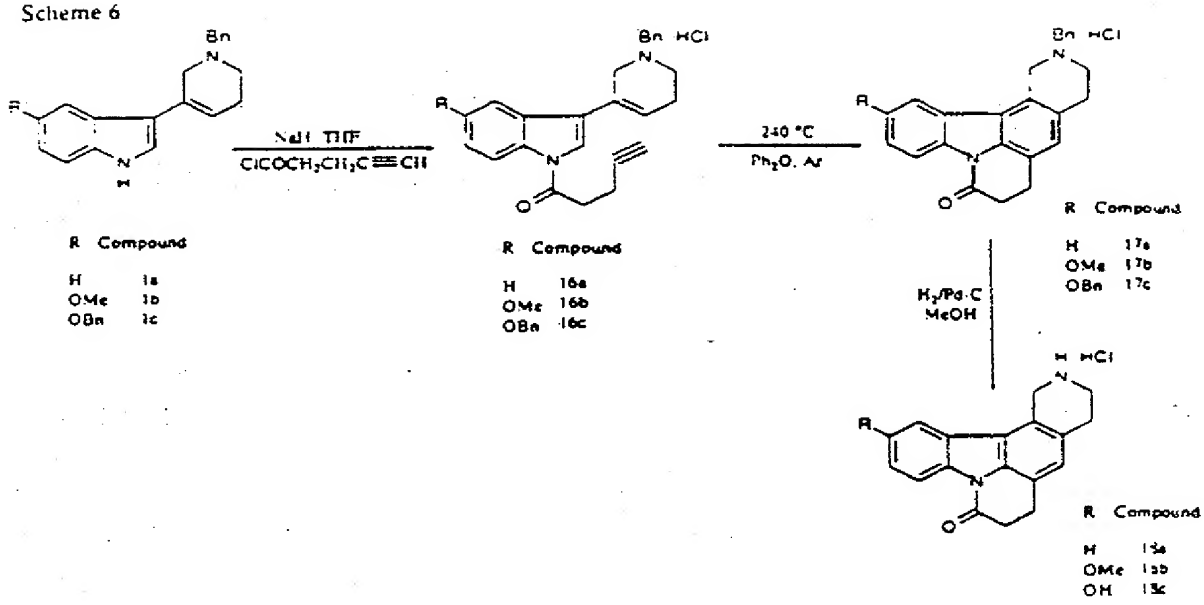
Scheme 4



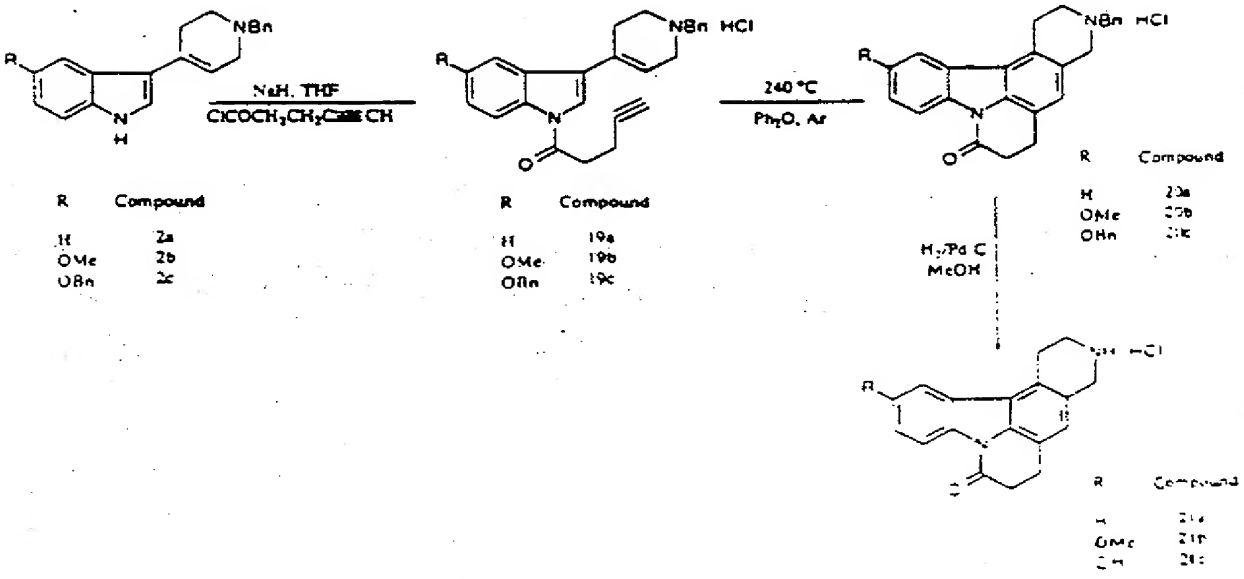
Scheme 5



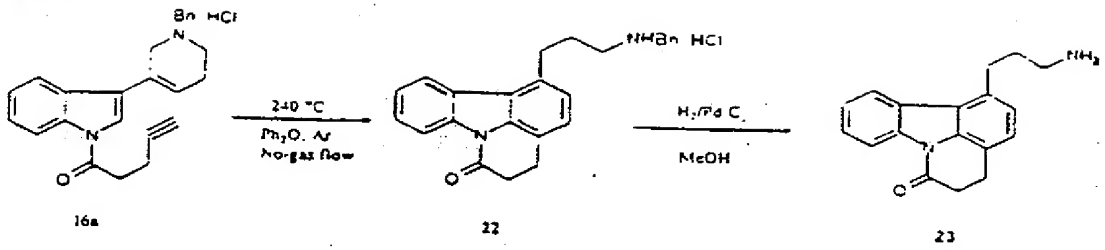
Scheme 6



Scheme 7

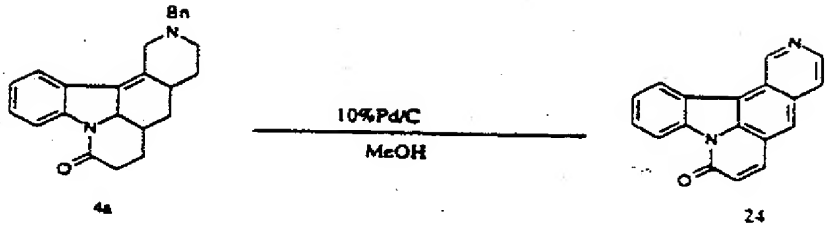


Scheme 8

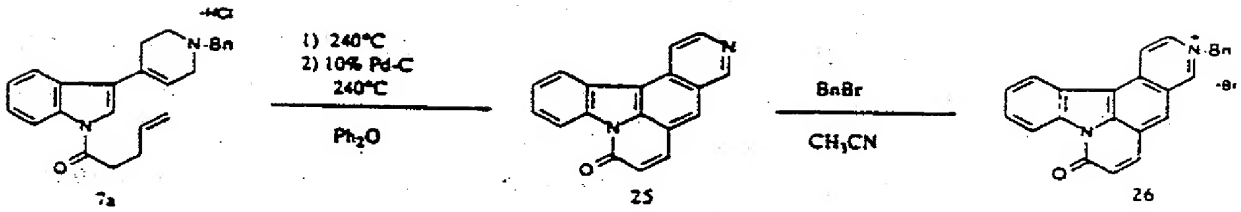


16a 22 23

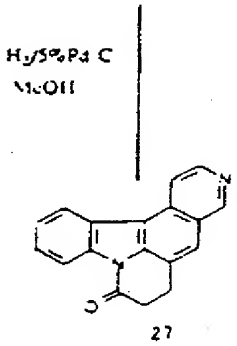
Scheme 9



4a 24



7a 25 26



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M&C FOLIO: 230P70859/FP-9414 WANGDOC: 1009D

EXAMPLE 1

(3aS,12R,12aS,12bR)-12-Amino-2,3,3a,4,11,12,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

1(a) (S)-1-(2-Cyclohexen-1-yl)acetyl-4-methoxy-1H-indol-3-carboxaldehyde

A solution of 5.47 g of 4-methoxy-1H-indol-3-carboxaldehyde in 624 ml of dimethoxyethane was added to a mixture of 63 ml of a 0.5 M solution of potassium bis(trimethylsilyl)amide in toluene with 31 ml of dimethoxyethane at -70°C. The resulting mixture was stirred for 20 minutes at this temperature, after which time 3.0 g of (S)-1-(2-cyclohexen-1-yl)acetyl chloride was added to the mixture, still at -70°C, and the reaction mixture was stirred for a further 30 minutes at the same temperature and then poured into a saturated aqueous solution of ammonium chloride and extracted with ethyl acetate. The extract was washed with brine, dried over anhydrous sodium sulfate, and evaporated under reduced pressure. The residue was purified by silica gel column chromatography using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent. The resulting product was reprecipitated from a mixture of methylene chloride and n-hexane to afford 6.07 g of the title compound, melting at 120-121°C.

Elemental analysis:

Calculated for $C_{18}H_{19}NO_3$:

C, 72.71%; H, 6.44%; N, 4.71%.

Found: C, 72.65%; H, 6.44%; N, 4.71%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1722, 1668.

Mass Spectrum (m/e):
297 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.32-1.45 (1H, multiplet),
1.55-1.82 (2H, multiplet),
1.88-2.09 (3H, multiplet),
2.81-2.98 (3H, multiplet),
4.04 (3H, singlet),
5.59-5.85 (2H, multiplet),
6.87 (1H, doublet, J=8Hz),
7.38 (1H, triplet, J=8Hz),
8.13 (1H, doublet, J=8Hz),
8.15 (1H, singlet),
10.57 (1H, singlet).

$[\alpha]_D^{23}$ +77.14° (c=1.12, trichloromethane).

1(b) (S)-Methyl (Z)-3[1-(2-cyclohexen-1-yl)acetyl-4-methoxy-1H-indol-3-yl]propenoate

34.5 ml of a solution of 0.5 M potassium bis(trimethylsilyl)amide in toluene was added to a mixture of 3.6 ml of bis(2,2,2-trifluoroethyl)(methoxy carbonylmethyl)phosphonate with 22.51 g of 18-crown-6 in 340 ml of tetrahydrofuran at -70°C. The resulting mixture was stirred for 20 minutes at this temperature, after which time a solution of 5.07 g of (S)-1-(2-cyclohexen-1-yl)acetyl-4-methoxy-1H-indol-3-carboxaldehyde, as obtained in Example 1(a), in 85 ml of tetrahydrofuran was added to the mixture at -70°C. The reaction mixture was then stirred for a further 30 minutes at this temperature, and then poured into a saturated aqueous

solution of ammonium chloride and extracted with ethyl acetate. The extract was washed with brine and dried over anhydrous sodium sulfate. The solvent was removed by distillation under reduced pressure, and the residue was purified by silica gel chromatography using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent. The resulting product was crystallized from cyclohexane to afford 6.00 g of the title compound, melting at 94°C.

Elemental analysis:

Calculated for $C_{21}H_{23}NO_4$:

C, 71.37%; H, 6.56%; N, 3.96%.

Found: C, 71.37%; H, 6.29%; N, 4.09%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

1713.

Mass Spectrum (m/e):

353 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.36-1.5 (1H, multiplet),
1.6-1.7 (1H, multiplet),
1.72-1.85 (1H, multiplet),
1.91-2.09 (3H, multiplet),
2.85-3.02 (3H, multiplet),
3.77 (3H, singlet),
3.95 (3H, singlet),
5.65-5.83 (2H, multiplet),
5.93 (1H, doublet, J=10Hz),
6.79 (1H, doublet, J=8Hz),
7.3 (1H, triplet, J=8Hz),
7.86 (1H, doublet, J=10Hz),
8. 17 (1H, doublet, J=8Hz),
9.02 (1H, singlet).

1(c) Methyl (3aS,12R,12aS,12bR,12cS)-1,2,3,3a,4,5,12,12a,12b,12c-decahydro-10-methoxy-5-oxo-isoquino[2,1,8-1ma]carbazol-12-carboxylate

0.5 g of (S)-methyl (Z)-3-[1-(2-cyclohexen-1-yl)-acetyl-4-methoxy-1H-indol-3-yl]propenoate, as obtained in Example 1(b), was added to 150 ml of biphenyl ether and the mixture was refluxed for 30 minutes. The reaction mixture was cooled to room temperature and purified by silica gel chromatography using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent. The resulting product was crystallized from hexane to afford 0.374 g of the title compound, melting at 185-187°C.

Elemental analysis:

Calculated for $C_{21}H_{23}NO_4 \cdot 1/3H_2O$:
C, 70.18%; H, 6.64%; N, 3.90%.
Found: C, 70.40%; H, 6.52%; N, 3.84%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1736, 1656.

Mass Spectrum (m/e):
353 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.32-1.72 (5H, multiplet),
1.82-1.9 (1H, multiplet),
2.1-2.22 (2H, multiplet),
2.37 (1H, doublet, J=32Hz),
2.41-2.56 (1H, multiplet),
2.82 (1H, doublet of doublets, J=8&20Hz),
3.73-3.82 (1H, multiplet),
3.77 (3H, singlet),
3.9 (3H, singlet),

4.75-4.83 (1H, multiplet),
6.06-6.11 (1H, multiplet),
6.64 (1H, doublet, J=8Hz),
7.22 (1H, triplet, J=8Hz),
7,75 (1H, doublet, J=8Hz).

1(d) Methyl (3aS,12R,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-methoxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylate

20 Drops of concentrated aqueous hydrochloric acid were added to a solution of 1.1 g of methyl (3aS,12R,12aS,12bR,12cS)-1,2,3,3a,4,5,12,12a,12b,12c-decahydro-10-methoxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylate, as obtained in Example 1(c), in 110 ml of ethanol under reflux conditions, and refluxing was continued for 20 minutes. The reaction mixture was then cooled to 0°C and the resulting crystals were collected by filtration to afford 0.988g of the title compound, melting at 230-231°C.

Elemental analysis:
Calculated for $C_{21}H_{23}NO_4 \cdot 1/5H_2O$:
C, 70.65%; H, 6.61%; N, 3.92%.
Found: C, 70.68%; H, 6.57%; N, 3.91%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1730, 1706.

Mass Spectrum (m/e):
353 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.8-1.7 (6H, multiplet),
2.2-2.3 (1H, multiplet),
2.48-2.69 (2H, multiplet),

2.88-3.28 (5H, multiplet),
3.76 (3H, singlet),
3.9 (3H, singlet),
6.71 (1H, doublet, J=8Hz),
7.2 (1H, doublet, J=8Hz),
8.0 (1H, doublet, J=8Hz).

1(e) (3aS,12R,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-Decahydro-10-methoxy-5-oxoisoquino[2,1,8-lma]-carbazol-12-carboxylic acid

8 ml of a 30% w/v aqueous solution of potassium hydroxide was added to a solution of 200 mg methyl (3aS,12R,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-methoxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylate, as obtained in Example 1 (d), in 80 ml of methanol and the mixture was refluxed for 15 minutes. The reaction mixture was cooled to 0°C and then ice, concentrated aqueous hydrochloric acid and dioxane were added. The reaction mixture was stirred for 1 hour at room temperature and then the solvent was removed by evaporation under reduced pressure. The resulting crystals were collected by filtration to afford 186 mg of the title compound, melting at >300°C.

Elemental analysis:
Calculated for $C_{20}H_{21}NO_4 \cdot 1/3H_2O$:
C, 69.55%; H, 6.32%; N, 4.06%.
Found: C, 69.76%; H, 6.68%; N, 4.13%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3215,1741,1683 cm^{-1}. Mass Spectrum (m/e):
339 (M^+).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
0.8-1.0 (1H, multiplet),

1.0-1.75 (5H, multiplet),
2.0-2.7 (5H, multiplet),
2.8-3.3 (4H, multiplet),
3.9 (3H, singlet),
6.7 (1H, doublet, J=8Hz),
7.2 (1H, triplet, J=8Hz),
7.95 (1H, doublet, J=8Hz).

1(f) (3aS,12R,12aS,12bR)-12-Benzyloxycarbonylamino-2,3,3a,4,11,12,12a,12b-octahydro-10-methoxyisoquino-[2,1,8-lma]carbazol-5(1H)-one

74 µl of triethylamine and 63 µl of ethyl chloroformate were added to a suspension of 150 mg of (3aS,12R,12aS,12bR)-1,2,3,3a,4,5,12,12a,12b-decahydro-10-methoxy-5-oxoisoquno[2,1,8-lma]carbazol-12-carboxylic acid, as obtained in Example 1 (e), in 3 ml of acetone at 0°C, and the resulting mixture was stirred for 15 minutes at this temperature. After this time, a solution of 43 mg of sodium azide in 0.5 ml of water was added to the reaction mixture, still at 0°C. The reaction mixture was then stirred for 1 hour at 0°C, poured into water, and extracted three times successively with ethyl acetate. The extract was washed with brine and dried over anhydrous sodium sulfate. The solvent was removed by evaporation under reduced pressure to afford the acid azide in the form of crystals. A suspension of these crystals in 2 ml of toluene was refluxed for 4 hours and 105 µl of benzyl alcohol was then added to this mixture. After refluxing the resulting mixture for a further 7 hours, the reaction mixture was cooled to room temperature, and 97 mg of the resulting crystals were collected by filtration, melting at >300°C.

Elemental analysis:

Calculated for $C_{27}H_{28}N_2O_4$:

C, 72.95%; H, 6.35%; N, 6.30%.
Found: C, 73.14%; H, 6.60%; N, 6.10%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3354, 1710, 1687.

Mass Spectrum (m/e):
444 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.8-1.4 (3H, multiplet),
1.5-1.8 (3H, multiplet),
2.1-2.4 (2H, multiplet),
2.48-2.57 (1H, multiplet),
2.62 (1H, doublet of doublets, J=2&17Hz),
2.9-3.0 (1H, multiplet),
3.2-3.4 (3H, multiplet),
3.86 (3H, singlet),
4.1-4.26 (1H, multiplet),
4.9-5.2 (3H, multiplet),
6.7 (1H, doublet, J=8Hz),
7.13-7.4 (6H, multiplet),
8.0 (1H, doublet, J=8Hz).

1(g) (3aS,12R,12aS,12bR)-12-Amino-2,3,3a,4,11,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

551 µl of a 1M solution of boron tribromide in methylene chloride was added to a suspension of 70 mg of (3aS,12R,12aS,12bR)-12-benzyloxycarbonylamino-2,3,3a,4,11,12,12a,12b-octahydro-10-methoxyisoquino[2,1,8-lma]-carbazol-5(1H)-one, as obtained in Example 1 (f), in 551 µl of methylene chloride at 0°C, and the resulting mixture was stirred for 24 hours after being allowed to warm to room temperature. After this time, 7 ml of

water and 5 ml of a saturated aqueous solution of sodium hydrogencarbonate were added to the reaction mixture which was then stirred for 0.5 hour at room temperature. The crystals which precipitated were collected by filtration, washed with water, and suspended in 551 µl of methanol. 551 µl of a 4N solution of hydrogen chloride in dioxane was added to the resulting suspension and the reaction mixture was then stirred at room temperature for 1 hour. After this time, 1 ml of isopropyl ether was added to the reaction mixture which was then stirred for a further 0.5 hour. 40 mg of the resulting crystals were collected by filtration, melting at >300°C.

Elemental analysis:

Calculated for $C_{18}H_{21}ClN_2O_2 \cdot 5/4H_2O$:
C, 60.84%; H, 6.67%; N, 7.88%; Cl, 9.98%.
Found: C, 60.87%; H, 6.59%; N, 7.66%; Cl, 9.57%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3215, 1715.

Mass Spectrum (m/e):
296 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
0.63-0.75 (1H, multiplet),
0.87-1.0 (1H, multiplet),
1.15-1.3 (1H, multiplet),
1.5-1.73 (3H, multiplet),
2.14-2.26 (1H, multiplet),
2.29-2.38 (1H, multiplet),
2.68-2.81 (1H, multiplet),
3.08 (1H, doublet of doublets, J=5&17Hz),
3.23-3.4 (4H, multiplet),
3.63-3.71 (1H, broad singlet),

6.67 (1H, doublet, J=8Hz),
7.03 (1H, triplet, J=8Hz),
7. 68 (1H, doublet, J=8Hz),
8.29-8.44 (2H, broad singlet)

$[\alpha]_D^{23}$ -60.9° (c=0.46, H_2O).

EXAMPLE 2

(3aS,12R,12aS,12bR)-12-Ethlyamino-2,3,3a,4,11,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

and

(3aS,12R,12aS,12bR)-12-Diethylamino-2,3,3a,4,11,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

2(a) (3aS,12R,12aS,12bR)-12-Amino-2,3,3a,4,11,12a,12b-octahydro-10-methoxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

0.55 g of the product of Example 1(f) and 0.55 g of 10% w/w palladium on carbon were added to a mixture consisting of 250 ml of dioxane, 50 ml of methanol, 50 ml of water and 0.5 ml of concentrated aqueous hydrochloric acid at 40°C. The resulting mixture was stirred for 6 hours under 1 atmosphere of hydrogen at the same temperature. After this time, the reaction mixture was filtered and the catalyst was washed with a warm solution of aqueous methanol. The water and ethanol were removed by evaporation under reduced pressure, and the residue was reprecipitated from a mixture of methanol and isopropyl ether to afford 0.285 g of the title compound, melting at >300°C.

Mass Spectrum (m/e):
310 (M^+-HCl).

2(b) (3aS,12R,12aS,12bR)-12-Ethylamino-2,3,3a,4,11,12a,12b-octahydro-10-methoxyisoquino-[2,1,8-lma]carbazol-5(1H)-one

and

2(c) (3aS,12R,12aS,12bR)-12-Diethylamino-2,3,3a,4,11,12a,12b-octahydro-10-methoxyisoquino-[2,1,8-lma]carbazol-5(1H)-one

231 µl of iodoethane and 121 mg of sodium hydrogencarbonate were added to a suspension of 0.1 g of the compound obtained in Example 2(a) in 5 ml of benzene, and the resulting mixture was stirred vigorously for 8 hours at 90°C in a pressure bottle. The reaction mixture was then poured into water and extracted with ethyl acetate. The extract was dried over anhydrous sodium sulfate and the solvent was removed by evaporation under reduced pressure. The residue was purified by silica gel chromatography using a 100 : 10 : 1 by volume mixture of methylene chloride, methanol and aqueous ammonia as the eluent to give each of the title compounds as an amorphous solid, each in a yield of 18 mg.

The compound of Example 2(b)

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.8-1.37 (3H, multiplet),
1.31 (3H, triplet, J=8Hz),
1.52-1.81 (3H, multiplet),
2.18-2.45 (2H, multiplet),
2.54-2.71 (1H, multiplet),

2.62 (1H, doublet of doublets, J=2&17Hz),
2.9-3.02 (3H, multiplet),
3.17-3.26 (1H, broad singlet),
3.29-3.43 (3H, multiplet),
3.91 (3H, singlet),
6.74 (1H, doublet, J=8Hz),
7.22 (1H, triplet, J=8Hz),
7.95 (1H, doublet, J=8Hz).

The compound of Example 2(c).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.82-1.37 (3H, multiplet),
1.19 (6H, triplet, J=7Hz),
1.51-1.84 (3H, multiplet),
2.14-2.42 (2H, multiplet),
2.62 (1H, doublet of doublets, J=2&17Hz),
2.8-3.09 (5H, multiplet),
3.11-3.32 (3H, multiplet),
3.9 (3H, singlet),
6.71 (1H, doublet, J=8Hz),
7.2 (1H, triplet, J=8Hz),
7.98 (1H, doublet, J=8Hz).

2(d) (3aS,12R,12aS,12bR)-12-Ethylamino-2,3,3a,4,11,-12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

186 µl of a 1M boron tribromide solution in methylene chloride was added to a solution of 18 mg of the compound obtained in Example 2(b) in 1 ml of dry methylene chloride at 0°C, and the resulting mixture was stirred overnight at room temperature. After this time 1 ml of water and 5 ml of a saturated aqueous solution of sodium hydrogencarbonate were added to the reaction mixture which was then stirred for 30 minutes. The

precipitated solid was filtered off, washed with water, and added to a mixture of 186 μl of water and 19 μl of 4N hydrogen chloride in dioxane. The reaction mixture was stirred for 1 hour and then stirred for a further 1 hour after the addition of 1 ml of isopropyl ether. 14 mg of the title compound was collected by filtration, melting at 151-154°C.

Mass Spectrum (m/e):
324 (M^+-HCl).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3163, 1678, 1637.

$[\alpha]_D^{23}$ -10.67° (c=0.15, dimethylsulfoxide).

2(e) (3aS,12R,12aS,12bR)-12-Diethylamino-2,3,3a,4,11,12a,12b-octahydro-10-hydroxyisoquino-[2,1,8-lma]carbazol-5(1H)-one hydrochloride

20 mg of the title compound was obtained from 18 mg of the compound obtained in Example 2(b) employing a similar procedure to that described in Example 2(d) and appropriate amounts of materials, the title compound melting at 108-110°C.

Mass Spectrum (m/e):
352 (M^+-HCl).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3167, 1682, 1646.

$[\alpha]_D^{23}$ -14.29° (c=0.35, dimethylsulfoxide).

EXAMPLE 3

(2aS,11R,11aS,11bR)-11-Amino-2,2a,3,4,10,11,11a,11b-octahydro-4-oxo-1H-cyclopent[de]indolo[3,2,1-ij]quinoline hydrochloride

and

(2aR,11S,11aR,11bS)-11-Amino-2,2a,3,4,10,11,11a,11b-octahydro-4-oxo-1H-cyclopent[de]indolo[3,2,1-ij]quinoline hydrochloride

3(a) 1-(2-Cyclopenten-1-yl)acetyl-4-methoxy-1H-indol-3-carboxaldehyde

Following a procedure similar to that described in Example 1(a), but using 175 mg of 4-methoxy-1H-indol-3-caboxaldehyde, 152 mg of 2-(2-cyclopenten-1-yl)acetyl chloride and 2.2 ml of a 0.5 M solution of potassium bis(trimethylsilyl)amide in 25 ml of dimethoxyethane as starting materials and other agents in appropriate amounts, 180 mg of the title compound was obtained as a white solid, melting at 104-105°C.

Elemental analysis:

Calculated for $C_{17}H_{17}NO_3$:

C, 72.07%; H, 6.05%; N, 4.94%.

Found: C, 72.23%; H, 6.01%; N, 5.24%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

1723, 1683.

Mass Spectrum (m/e):

283 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.45-1.62 (1H, multiplet),
2.17-2.55 (3H, multiplet),
2.83-3.06 (2H, multiplet),
3.19-3,37 (1H, multiplet),
3.98 (3H, singlet),
5.67-5.84 (2H, multiplet),
6.79 (1H, doublet, J=8Hz),
7.27 (1H, triplet, J=8Hz),
8.06 (1H, doublet, J=8Hz),
8.07 (1H, singlet),
10.47 (1H, singlet).

3(b) Methyl (Z)-3-[1-(2-cyclopenten-1-yl)acetyl-4-methoxy-1H-indol-3-yl]propenoate

Following a procedure similar to that described in Example 1(b), but using 1.83 g of the compound obtained in Example 3(a), 1.4 ml of bis(2,2,2-trifluoroethyl)-(methoxycarbonylmethyl)phosphonate and a mixture of 12.9 ml of a 0.5 M solution of potassium bis(trimethylsilyl)amide in toluene and 8.53 g of 18-crown-6 in 161 ml of tetrahydrofuran as starting materials and other agents in appropriate amounts, 2.14 g of the title compound was obtained as a white solid, melting at 99-100°C.

Elemental analysis:

Calculated for $C_{20}H_{12}NO_4$:
C, 70.78%; H, 6.24%; N, 4.13%.
Found: C, 70.78%; H, 6.38%; N, 4.14%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1714.

Mass Spectrum (m/e):

339 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.57-1.69 (1H, multiplet),
2.21-2.48 (3H, multiplet),
2.94-3.15 (2H, multiplet),
3.29-3.43 (1H, multiplet),
3.77 (3H, singlet),
3.95 (3H, singlet),
5.77-5.86 (2H, multiplet),
5.93 (1H, doublet, J=10Hz),
6.78 (1H, doublet, J=8Hz),
7.3 (1H, triplet, J=8Hz),
7.87 (1H, doublet, J=10Hz),
8.16 (1H, doublet, J=8Hz),
9.0 (1H, singlet).

3(c) Methyl (2aα,11β,11aα,11bα,11cβ)-2,2a,3,4,11,11a,11b,11c-octahydro-4-oxo-1H-cyclopent[de]-indolo[3,2,1-ij]quinolin-11-carboxylate

A solution of 2.13 g of the compound obtained in Example 3(b) in 20 ml of mesitylene was refluxed for 8 hours, after which time the solvent was removed by evaporation under reduced pressure. The residue was chromatographed on silica gel using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent, and the resulting material was reprecipitated from a mixture of cyclohexane and methylene chloride. 2.09 g of the title compound was obtained as a white solid, melting at 182-183°C.

Elemental analysis:

Calculated for $C_{20}H_{21}NO_4$:

C, 70.78%; H, 6.24%; N, 4.13%.
Found: C, 70.42%; H, 6.22%; N, 4.14%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1728, 1676.

Mass Spectrum (m/e):
339 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.49-1.76 (2H, multiplet),
1.79-1.9 (1H, multiplet),
2.1-2.25 (2H, multiplet),
2.31-2.45 (2H, multiplet),
2.48-2.61 (1H, multiplet),
2.82 (1H, doublet of doublets, J=8&8Hz),
2.95-3.0 (1H, multiplet),
3.81 (3H, singlet),
3.92 (3H, singlet),
3.92-4.01 (1H, multiplet),
6.28-6.31 (1H, multiplet),
6.62 (1H, doublet, J=8Hz),
7.27 (1H, triplet, J=8Hz),
7.68 (1H, doublet, J=8Hz).

3(d) (2aα,11β,11aα,11bα,11cβ)-2,2a,3,4,11,11a,11b,11c-Octahydro-4-oxo-1H-cyclopent[de]-indolo[3,2,1-ij]quinolin-11-carboxylic acid

40 ml of a 30% w/v aqueous solution of potassium hydroxide was added to a solution of 853 mg of the compound obtained in Example 3 (c) in 200 ml of methanol, and the resulting mixture was refluxed for 15 minutes. The reaction mixture was cooled using an ice bath, after which ice and concentrated aqueous hydrochloric acid were added. Shortly after the

reaction mixture had been acidified, the solvent was removed by evaporation under reduced pressure, and the crystals which precipitated out were collected by filtration to yield 797 mg of the title compound.

Elemental analysis:

Calculated for $C_{19}H_{19}NO_4$:

C, 70.14%; H, 5.89%; N, 4.31%.

Found: C, 70.01%; H, 5.93%; N, 4.28%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

1702, 1675.

Mass Spectrum (m/e):

325 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.57-2.65 (9H, multiplet),
2.81-2.94 (1H, multiplet),
2.98-3.05 (1H, multiplet),
3.92-4.1 (1H, multiplet),
3.95 (3H, singlet),
6.43 (1H, singlet),
6.69 (1H, doublet, J=8Hz),
7.3 (1H, triplet, J=8Hz),
7 .71 (1H, doublet, J=8Hz).

3(e) (2aα,11β,11aα,11bα)-2,2a,3,4,10,11,11a,11b-Octahydro-4-oxo-1H-cyclopent[de]indolo[3,2,1-ij]quinolin-11-carboxylic acid

1 ml of 4N hydrogen chloride in dioxane was added to a solution of 70 mg of the compound obtained in Example 3(d) in 1 ml of dioxane and the resulting mixture was stirred for 1 hour. Subsequently, the solvent was removed by evaporation under reduced pressure to give

70 mg of the title compound.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
0.98-1.19 (1H, multiplet),
1.36-1.51 (1H, multiplet),
1.77-1.96 (2H, multiplet),
2.23-2.5 (1H, broad singlet),
2.53-2.63 (1H, multiplet),
2.84 (1H, doublet, J=12Hz),
2.9-3.15 (4H, multiplet),
3.18-3.26 (1H, multiplet),
3.5 (1H, doublet, J=12Hz),
3.91 (3H, singlet),
6.73 (1H, doublet, J=8Hz),
7.22 (1H, triplet, J=8Hz),
7.97 (1H, doublet, J=8Hz).

3(f) (2aS,11R,11aS,11bR)-11-Amino-2,2a,3,4,10,11,11a,11b-octahydro-4-oxo-1H-cyclopent[de]indolo[3,2,1-ij]quinoline hydrochloride

and

3(g) (2aR,11S,11aR,11bS)-11-Amino-2,2a,3,4,10,11,11a,11b-octahydro-4-oxo-1H-cyclopent[de]indolo[3,2,1-ij]quinoline hydrochloride

(i) A procedure similar to that described in Example 1(f) was followed, but using, as starting materials, 100 mg of the compound obtained in Example 3(e) in 5 ml of acetone, 44 μl of ethyl chloroformate, 52 μl of triethylamine and 30 mg of sodium azide in 100 μl of water. Other agents were used in the appropriate amounts. A solution of the resulting acid azide and 37 μl of (R)-phenethyl alcohol in 2 ml of toluene was refluxed for 6 hours to give 2 diastereomers of the

carbamate compound. The less polar isomer was separated by Lobar chromatography (column size B, LiChroprep Si 60), using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent. The resulting compound was treated with a preparation of 1M boron tribromide in methylene chloride (236 µl in 236 µl), following a procedure similar to that of Example 1(g), to afford 3 mg of one of the enantiomers having the following optical rotation characteristics: $[\alpha]_D^{23}$ +70° (c=0.1, H_2O).

(ii) The other enantiomer was obtained as follows. The mixture of the 2 diastereomers was first treated with boron tribromide in a similar fashion to that described in (i) above, to afford 19 mg of a mixture of enantiomers, which were separated by HPLC (column:CHIRAL CEL OD), using a 95 : 5 by volume mixture of hexane and isopropanol as the eluent, at a flowrate of 1 ml/min, to yield 1 mg of each of the two enantiomers respectively.

Retention times:
the less polar compound: 22 minutes
the more polar compound: 28 minutes

The mixture of the two enantiomers had the following properties:

Melting point >300°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3310, 1716, 1682.

Mass Spectrum (m/e):
282 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:

0.83-0.95 (1H, multiplet),
1.27-1.39 (1H, multiplet),
2.47-2.59 (2H, multiplet),
2.63 (1H, doublet, J=8Hz),
2.67-2.77 (2H, multiplet),
3.07 (1H, doublet, J=8Hz),
3.16-3.27 (4H, multiplet),
3.63-3.77 (1H, broad singlet),
6.7 (1H, doublet, J=8 Hz),
7.11 (1H, triplet, J=8Hz),
7.67 (1H, doublet, J=8Hz).

Example 4

(2S,3aR)-2-Amino-1,2,3,3a,4,5-hexahydro-6H-pyrido-[3,2,1-jk]carbazol-6-one hydrochloride

and

(2R,3aS)-2-Amino-1,2,3,3a,4,5-hexahydro-6H-pyrido-[3,2,1-jk]carbazol-6-one hydrochloride

4(a) 1-(4-Pentenoyl)-1H-indol-3-carboxaldehyde

Following a procedure similar to that of Example 1(a), but using 2.9 g of indol-3-caboxaldehyde as starting material, and other agents in appropriate amounts, 4.17 g of the title compound was obtained, melting at 103-104°C.

Elemental analysis:

Calculated for $C_{14}H_{13}NO_2$:
C, 73.99%; H, 5.77%; N, 6.16%.
Found: C, 74.11%; H, 5.72%; N, 6.17%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1720, 1675.

Mass Spectrum (m/e):
227 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
2.83 (2H, quartet, J=7Hz),
3.09 (2H, triplet, J=7Hz),
5.09 (1H, doublet, J=10Hz),
5.15 (1H, doublet, J=17Hz),
5.83-6.01 (1H, multiplet),
7.39 (1H, triplet, J=8Hz),
7.44 (1H, triplet, J=8Hz),
8.08 (1H, singlet),
8.26 (1H, doublet, J=8Hz),
8.4 (1H, doublet, J=8Hz),
10.1 (1H, singlet).

4(b) Methyl (Z)-3-[1-(4-pentenoyl)-1H-indol-3-yl]-2-propenoate

Following a procedure similar to that described in Example 1(b), but using a solution of 1.2 g of the compound obtained in Example 4(a), as the starting material, and other agents in appropriate amounts, 1.46 g of the title compound was obtained, melting at 79-80°C.

Elemental analysis:
Calculated for $C_{17}H_{17}NO_3$:
C, 72.07%; N, 6.05%; N, 4.94%.
Found: C, 72.20%; H, 6.15%; N, 4.75%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1710.

Mass Spectrum (m/e):

283 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

2.65 (2H, quartet, J=7Hz),
3.18 (2H, triplet, J=7Hz),
3.8 (3H, singlet),
5.11 (1H, doublet, J=10Hz),
5.2 (1H, doublet, J=17Hz),
5.91-6.03 (1H, multiplet),
6.05 (1H, doublet, J=10Hz),
7.18 (1H, doublet, J=10Hz),
7.37 (1H, triplet, J=8Hz),
7.41 (1H, triplet, J=8Hz),
7.66 (1H, doublet, J=8Hz),
8.53 (1H, doublet, J=8Hz),
9.12 (1H, singlet).

4(c) Methyl (2α,3aβ,11cα)-2,3,3a,5,6,11c-hexahydro-6-oxo-4H-pyrido[3,2,1-jk]carbazol-2-carboxylate

A suspension of 5.71 g of the compound obtained in Example 4(b) in 50 ml of mesitylene was refluxed for 4 days, and the title compound was obtained following a similar purification procedure as that described in Example 3(c), in a yield of 3.91 g, melting at 137-139°C.

Elemental analysis:

Calculated for $C_{17}H_{17}NO_3$:
C, 72.07%; H, 6.05%; N, 4.94%.
Found: C, 71.80%; H, 6.16%; N, 4.90%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

1736, 1647.

Mass Spectrum (m/e):
283 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.61-2.14 (4H, multiplet),
2.13-2.31 (1H, multiplet),
2.55-2.81 (2H, multiplet),
3.57-3.69 (1H, multiplet),
3.79 (3H, singlet),
4.14-4.21 (1H, multiplet),
5.88-5.91 (1H, multiplet),
7.09 (1H, triplet, J=8Hz),
7.29 (1H, triplet, J=8Hz),
7.43 (1H, doublet, J=8Hz),
8.1 (1H, doublet, J=8 Hz).

4(d) Methyl (2α,3aβ)-2,3,3a,4,5,6-hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-carboxylate

A suspension of 2 g of the compound obtained in Example 4(c) and 1 g of 10% w/w palladium on carbon in 100 ml of ethanol was refluxed for 0.5 hour. The catalyst was subsequently filtered off and the solvent was removed by evaporation under reduced pressure to afford 1.9 g of the title compound, melting at 125-127°C.

Elemental analysis:
Calculated for $C_{17}H_{17}NO_3$:
C, 72.07%; H, 6.05%; N, 4.94%.
Found: C, 72.27%; H, 6.11%; N, 4.93%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1738, 1791.

Mass Spectrum (m/e):
283 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.55-1.79 (1H, multiplet),
2.14-2.22 (1H, multiplet),
2.43-2.52 (1H, multiplet),
2.71-3.12 (6H, multiplet),
3.79 (3H, singlet),
7.21-7.34 (2H, multiplet),
7.41 (1H, doublet, J=8Hz),
8.39 (1H, doublet, J=8Hz).

4(e) (2α, 3aβ)-2,3,3a,4,5,6-Hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-carboxylic acid

Following a procedure similar to that of Example 1(e), but using 900 mg of the compound obtained in Example 4(d), and other agents in appropriate amounts, 738 mg of the title compound was obtained, melting at 274-275°C.

Elemental analysis:

Calculated for $C_{16}H_{15}NO_3$:
C, 71.36%; H, 5.61%; N, 5.20%.
Found: C, 71.27%; H, 5.66%; N, 5.24%.

Mass Spectrum (m/e):

269 (M^+).

4(f) (1S)-Phenethyl-N-[(2S,3aR)-2,3,3a,4,5,6-hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-yl]carbamate

and

4(g) (1S)-Phenethyl-N-[(2R,3aS)-2,3,3a,4,5,6-hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-yl]carbamate

Following a procedure similar to that of Example

3(g), but using 272 mg of the compound obtained in Example 4(e), a mixture of the title compounds was obtained. The mixture was purified by Lobar chromatography using a 1 : 3 by volume mixture of cyclohexane and ethyl acetate as the eluent to afford each of the title compounds.

The less polar compound was obtained in a yield of 160 mg, melting at 149-151°C.

$[\alpha]_D^{23}$ -86.79° (c=0.84, trichloromethane).

Elemental analysis:

Calculated for $C_{24}H_{24}N_2O_3 \cdot 1/2H_2O$:
C, 72.52%; H, 6.34%; N, 7.07%.
Found: C, 72.82%; H, 6.45%; N, 6.75%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1698, 1688.

Mass Spectrum (m/e):
388 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.55-1.75 (1H, multiplet),
1.56 (3H, singlet),
2.08-2.09 (1H, multiplet),
2.31-2.42 (2H, multiplet),
2.76-2.82 (2H, multiplet),
2.94-3.09 (1H, multiplet),
3.17-3.26 (1H, multiplet),
4.09-4.23 (1H, multiplet),
4.81-4.9 (1H, multiplet),
5.79-5.91 (1H, multiplet),
7.26-7.4 (9H, multiplet),
8.37 (1H, doublet, J=8Hz).

The more polar compound was obtained in a yield of 150 mg, melting at 192-193°C.

$[\alpha]_D^{23}$ +37.43° (c=1.05, trichloromethane).

Elemental analysis:

Calculated for $C_{24}H_{24}N_2O_3$:

C, 74.21%; H, 6.23%; N, 7.21%.

Found: C, 74.23%; H, 6.30%; N, 7.24%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

1699, 1686.

Mass Spectrum (m/e):

388 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.55-1.86 (1H, multiplet),
1.58 (3H, singlet),
2.12-2.21 (1H, multiplet),
2.31-2.49 (2H, multiplet),
2.75-2.82 (2H, multiplet),
2.96-3.21 (2H, multiplet),
4.07-4.23 (1H, multiplet),
4.81-4.9 (1H, multiplet),
5.79-5.91 (1H, multiplet),
7.26-7.4 (9H, multiplet),
8.37 (1H, doublet, J=8Hz).

4(h) (2S,3aR)-2-Amino-1,2,3,3a,4,5-hexahydro-6H-pyrido-[3,2,1-jk]carbazol-6-one hydorochloride

and

4(i) (2R,3aS)-2-Amino-1,2,3,3a,4,5-hexahydro-6H-pyrido-[3,2,1-jk]carbazol-6-one hydorochloride

A suspension of 160 mg of the less polar compound obtained in Example 4(f) and 100 mg of 10% w/w palladium on carbon in 10 ml of ethanol containing 1 ml of concentrated aqueous hydrochloric acid was stirred under a hydrogen atmosphere for 3 hours. After this time, the catalyst was filtered off and the solvent was removed by evaporation under reduced pressure. The residual solid was reprecipitated from a mixture of methanol, ethyl acetate and isopropyl ethyl ether to afford 62 mg of one enantiomer. Following a similar procedure, but starting with 150 mg of the more polar compound, 92 mg of the other enantiomer was obtained.

The properties of the enantiomer obtained from the less polar compound obtained in Example 4(f) were as follows:

Melting point: 279-281°C.

$[\alpha]_D^{23}$ -103.3° (c=0.09, H_2O).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3448, 1693.

Mass Spectrum (m/e):
240 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
1.57-1.77 (2H, multiplet),

2.1-2.17 (1H, multiplet),
2.35-2.4 (1H, multiplet),
2.61-2.74 (2H, multiplet),
2.86-2.96 (1H, multiplet),
3.05-3.15 (2H, multiplet),
3.6-3.72 (1H, broad singlet),
7.25-7.33 (2H, multiplet),
7.47-7.52 (1H, multiplet),
8.22-8.2 7 (1H, multiplet),
8.35-8.55 (2H, broad singlet).

The properties of the enantiomer obtained from the more polar compound obtained in Example 4(f) were as follows:

Melting point: 263-265°C.

$[\alpha]_D^{23}$ +110° (c=0.11, H_2O).

EXAMPLE 5

(2α,3aβ)-2-Aminomethyl-1,2,3,3a,4,5-hexahydro-6H-pyrido[3,2,1-jk]carbazol-6-one hydrochloride

5(a) (2α,3aβ)-2-Hydroxymethyl-1,2,3,3a,4,5-hexahydro-6H-pyrido[3,2,1-jk]carbazol-6-one

742 µl of a 1M solution of diborane complexed in tetrahydrofuran was added to a solution of 0.1 g of the compound obtained in Example 4(e) in 5 ml of tetrahydrofuran, at 0°C. The reaction mixture was stirred for 2 hours at room temperature, poured into water, and extracted with ethyl acetate. After drying over anhydrous sodium sulfate, the solvent was removed by evaporation under reduced pressure and the residue was chromatographed on silica gel using a 1 : 3 by volume mixture of cyclohexane and ethyl acetate as the

eluent, to give 71 mg of the title compound as a solid, melting at 152-154°C.

Elemental analysis:

Calculated for $C_{16}H_{17}NO_2$:

C, 75.27%; H, 6.71%; N, 5.49%.

Found: C, 75.11%; H, 6.89%; N, 5.34%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

3531, 1689, 1635.

Mass Spectrum (m/e):

255 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.21-1.41 (1H, multiplet),
1.65-1.82 (1H, multiplet),
2.19-2.47 (4H, multiplet),
2.5-3.09 (5H, multiplet),
3.7-3.85 (2H, multiplet),
7.28-7.4 (2H, multiplet),
7.47 (1H, doublet, J=8Hz),
8.42 (1H, doublet, J=8Hz).

5(b) (2α,3aβ)-2-Methanesulfonyloxymethyl-1,2,3,3a,4,5-hexahydro-6H-pyrido[3,2,1-jk]carbazol-6-one

A mixture of 237 mg of the compound obtained in Example 5(a), 108 μl of methansulfonyl chloride and 3 ml of pyridine was stirred for 3 hours. After this time, the reaction mixture was poured into water, and extracted with ethyl acetate. The extract was dried over anhydrous sodium sulfate and the solvent was removed by evaporation under reduced pressure. The resulting residue was then purified by silica gel chromatography, using a 1 : 1 by volume mixture of

cyclohexane and ethyl acetate as the eluent, to afford 250 mg of the title compound.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1701, 1636.

Mass Spectrum (m/e):
333 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.3-1.42 (1H, multiplet),
1.6-1.82 (1H, multiplet),
2.12-2.55 (4H, multiplet),
2.74-3.0 (4H, multiplet),
3.1 (3H, singlet),
4.15-4.4 (2H, multiplet),
7.23-7.32 (1H, multiplet),
7.41 (1H, doublet, J=8Hz),
8.39 (1H, doublet, J=8Hz).

5(c) (2α,3aβ)-2-Azidomethyl-1,2,3,3a,4,5-hexahydro-6H-pyrido[3,2,1-jk]carbazole-6-one

A suspension of 240 mg of the compound obtained in Example 5(b), 143 ml of trimethylsilyl azide, 454 mg of tetrabutyl ammonium fluoride, and a catalytic amount of molecular sieve 4A in 10 ml of tetrahydrofuran was refluxed for 2 hours. After removing the molecular sieve by filtration, the mixture was poured into water and extracted with ethyl acetate. The extract was dried over anhydrous sodium sulfate and the solvent was removed by evaporation under reduced pressure. The resulting residue was chromatographed on silica gel, using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent, to afford 170 mg of the title compound as a white solid.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
2100, 1701, 1637.

Mass Spectrum (m/e):
280 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.3-1.42 (1H, multiplet),
1.69-1.91 (1H, multiplet),
2.2-2.49 (4H, multiplet),
2.81-3.1 (4H, multiplet),
3.47-3.61 (2H, multiplet),
7.3-7.4 (2H, multiplet),
7.42-7.51 (1H, multiplet),
8.42-8.48 (1H, multiplet).

5(d) (2α,3aβ)-2-Aminomethyl-1,2,3,3a,4,5-hexahydro-6H-pyrido[3,2,1-jk]carbazol-6-one hydrochloride

A suspension of 170 mg of the compound obtained in Example 5(c) and 170 mg of 10% w/w palladium on carbon in 10 ml of ethanol containing 1 ml of concentrated aqueous hydrochloric acid was stirred under a hydrogen atmosphere for 3 hours. After this time, the catalyst was removed by filtration, and the solvent was removed by evaporation under reduced pressure. The resulting residue was reprecipitated from a mixture of methanol and ethyl acetate to afford 131 mg of the title compound, melting at 257-259°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3380, 1715, 1699.

Mass Spectrum (m/e):
254 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
1.07-1.14 (1H, multiplet),
1.78-1.87 (1H, multiplet),
2.02-2.07 (1H, multiplet),
2.09-2.2 (3H, multiplet),
2.82-2.87 (1H, multiplet),
2.9-3.03 (6H, multiplet),
7.13-7.16 (2H, multiplet),
7.18-7.23 (1H, multiplet),
8.05-8.15 (3H, multiplet).

EXAMPLE 6

(2α,3aβ,11bα,11cα)-2-Amino-1,2,3,3a,4,5,11b,11c-octahydro-6H-pyrido[3,2,1-jk]carbazol-6-one hydrochloride

and

(2α,3aβ,11bβ,11cα)-2-Amino-1,2,3,3a,4,5,11b,11c-octahydro-6H-pyrido[3,2,1-jk]carbazol-6-one hydrochloride

6(a) Methyl (2α,3aβ,11bα,11cα)-2,3,3a,4,5,6,11b,11c-octahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-carboxylate

and

6(b) Methyl (2α,3aβ,11bβ,11cα)-2,3,3a,4,5,6,11b,11c-octahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-carboxylate

A suspension of 1.5 g of the compound obtained in Example 4(c) and 0.1 g of 10% w/w palladium on carbon in a mixture of 25 ml each of tetrahydrofuran and ethanol

was stirred under a hydrogen atmosphere for one day. After this time, the catalyst was removed by filtration, and the solvent was removed by evaporation under reduced pressure. The resulting residue was purified by Lobar chromatography using a 1 : 3 by volume mixture of cyclohexane and ethyl acetate as the eluent, to afford the title compounds separately.

The less polar compound was obtained in a yield of 520 mg, melting at 160-161°C.

Elemental analysis:

Calculated for $C_{17}H_{19}NO_3$:
C, 71.56%; H, 6.71%; N, 4.91%.
Found: C, 71.41%; H, 6.72%; N, 4.94%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1738, 1649.

Mass Spectrum (m/e):
285 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.4-1.66 (3H, multiplet),
1.9-2.11 (3H, multiplet),
2.13-2,25 (1H, multiplet),
2.48-2.61 (1H, multiplet),
2.65-2.88 (2H, multiplet),
3.52-3.63 (1H, multiplet),
3.73 (3H, singlet),
3.82 (1H, triplet, J=8Hz),
7.04 (1H, triplet, J=8Hz),
7.15-7.23 (2H, multiplet),
8.09 (1H, doublet, J=8Hz).

The more polar compound was obtained in a yield of 470 mg, melting at 176-178°C.

Elemental analysis:

Calculated for $C_{17}H_{19}NO_3$:

C, 71.56%; H, 6.71%; N, 4.91%.

Found: C, 71.54%; H, 6.78%; N, 4.99%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

1733, 1648.

Mass Spectrum (m/e):

285 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.4-1.67 (2H, multiplet),
1.68-2.05 (3H, multiplet),
2.24-2.33 (1H, multiplet),
2.56-2.76 (4H, multiplet),
2.79-2.91 (1H, multiplet),
3.13 (1H, triplet, J=10Hz),
3.74 (3H, singlet),
7.08 (1H, triplet, J=8Hz),
7.15-7.25 (2H, multiplet),
8.04 (1H, doublet, J=8Hz).

6(c) (2α,3aβ,11bα,11cα)-2,3,3a,4,5,6,11b,11c-Octahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-carboxylic acid

A mixture consisting of the whole of the less polar compound obtained in Example 6(a) together with 9 ml of 1N aqueous sodium hydroxide was stirred at room temperature for 3 hours. After this time, the reaction mixture was acidified with 1N aqueous hydrochloric acid at 0°C, and the solution was concentrated under reduced

pressure. The precipitated crystals were collected by filtration to afford 454 mg of the title compound.

Elemental analysis:
Calculated for $C_{16}H_{17}NO_3 \cdot 1/4H_2O$:
C, 69.67%; H, 6.39%; N, 5.08%.
Found: C, 69.56%; H, 6.21%; N, 4.93%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3183, 1736, 1631.

Mass Spectrum (m/e):
271 (M^+).

6(d) (2α,3aβ,11bβ,11cα)-2,3,3a,4,5,6,11b,11c-Octahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-carboxylic acid

Following a procedure similar to that of Example 6(c), 373 mg of the title compound was obtained from 430 mg of the more polar compound obtained in Example 6(a).

Elemental analysis:
Calculated for $C_{16}H_{17}NO_3$:
C, 70.83%; H, 6.32%; N, 5.16%.
Found: C, 70.52%; H, 6.33%; N, 5.08%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3210, 1732, 1634.

Mass Spectrum (m/e):
271 (M^+).

6(e) Benzyl-N-[(2α,3aβ,11bα,11cα)-2,3,3a,4,5,6,11b,11c-octahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-yl]-carbamate

Following a procedure similar to that of Example 1(f), but using 400mg of the compound obtained in Example 6(c), and other agents in appropriate amounts, 510 mg of the title compound was obtained.

Elemental analysis:

Calculated for $C_{23}H_{24}N_2O_3 \cdot 1/3H_2O$:
C, 72.23%; H, 6.50%; N, 7.32%.
Found: C, 72.39%; H, 6.23%; N, 7.32%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1717, 1655.

Mass Spectrum (m/e):
376 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.85-1.04 (1H, multiplet),
1.4-1.6 (2H, multiplet),
1.84-2.03 (2H, multiplet),
2.1-2.34 (2H, multiplet),
2.47-2.8 (2H, multiplet),
3.53-3.65 (1H, multiplet),
3.8 (1H, triplet, J=10Hz),
3.85-4.05 (1H, multiplet),
4.65-4.84 (1H, broad singlet),
5.12 (2H, singlet),
7.06 (1H, triplet, J=8Hz),
7.12-7.43 (7 H, multiplet),
8.1 (1H, doublet, J=8Hz).

6(f) Benzyl-N-[(2α,3aβ,11bβ,11cα)-2,3,3a,4,5,6,11b,11c-octahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-yl]-carbamate

Following a procedure similar to that of Example 1(f), but using 300mg of the compound obtained in Example 6(d), and other agents in appropriate amounts, 400 mg of the title compound was obtained.

Elemental analysis:

Calculated for $C_{23}H_{24}N_2O_3 \cdot 1/4H_2O$:
C, 72.52%; H, 6.48%; N, 7.35%.
Found: C, 72.47%; H, 6.50%; N, 7.29%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1717, 1645.

Mass Spectrum (m/e):
376 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
1.1-1.3 (1H, multiplet),
1.39-1.6 (2H, multiplet),
1.9-2.05 (2H, multiplet),
2.26-2.38 (1H, multiplet),
2.57-2.75 (3H, multiplet),
2.85-3.01 (1H, multiplet),
3.12 (1H, triplet, J=10Hz),
3.8-4.02 (1H, broad singlet),
4.74-4.84 (1H, multiplet),
5.14 (2H, singlet),
7.08 (1H, triplet, J=8Hz),
7.12-7.4 (7H, multiplet),
8.02 (1H, doublet, J=8Hz).

6(g) (2α,3aβ,11bα,11cα)-2-Amino-1,2,3,3a,4,5,11b,11c-octahydro-6H-pyrido[3,2,1-jk]carbazol-6-one hydrochloride

Following a procedure similar to that of Example 5(d), but using 480mg of the compound obtained in Example 6(e), and other agents in appropriate amounts, 353 mg of the title compound was obtained, melting at 155-157°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3402, 1728, 1630.

Mass Spectrum (m/e):
242 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
1.4-1.5 (2H, multiplet),
1.8-2.4 (6H, multiplet),
2.66-2.91 (2H, multiplet),
3.3-3.42 (1H, multiplet),
3.7-3.81 (1H, multiplet),
3.95 (1H, triplet, J=10Hz),
6.95-7.1 (2H, multiplet),
7.13-7.22 (2H, multiplet),
7.9 (1H, doublet, J=8Hz).

6(h) (2α,3aβ,11bβ,11cα)-2-Amino-1,2,3,3a,4,5,11b,11c-octahydro-6H-pyrido[3,2,1-jk]carbazol-6-one hydrochloride

Following a procedure similar to that of Example 5(d), but using 340 mg of the compound obtained in Example 6(f), and other agents in appropriate amounts, 248 mg of the title compound was obtained, melting at 282-285°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3421, 1718, 1654.

Mass Spectrum (m/e):
242 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
1.25-1.7 (3H, multiplet),
1.86-2.29 (3H, multiplet),
2.38-2.68 (3H, multiplet),
2.8-3.18 (2H, multiplet),
3.26 (1H, triplet, J=10Hz),
3.25-3.49 (2H, multiplet),
6.96-7.11 (1H, multiplet),
7.15-7.31 (2H, multiplet),
7.85 (1H, doublet, J=8Hz).

EXAMPLE 7

7-1

(3aR,12R,12aR,12bS)-1,2,3,3a,4,5,11,12,12a,12b-Decahydro-10-hydroxy-5-oxoisoquino[2,1,8-1ma]carbazol-12-carboxylic acid

A suspension consisting of 150 mg of (3aR,12R,12aR, 12bS)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-benzyloxy-5-oxoisoquino[2,1,8-1ma]carbazol-12-carboxylic acid and 150 mg of 10% w/w palladium on carbon in a mixture of 3 ml of ethanol, 88 µl of pyridine and 5 ml of a 1N solution of aqueous sodium hydroxide was stirred for 3 hours under a hydrogen atmosphere. After this time, the catalyst was removed by filtration, the solution was acidified with 1N aqueous hydrochloric acid and then stirred for a further 1 hour. The reaction mixture was

concentrated under reduced pressure, and the precipitated crystals were collected by filtlation to afford 92 mg of the title compound, melting at 284-287°C.

$[\alpha]_D^{23}$ +92.05° (c=0.73, dimethylsulfoxide).

Elemental analysis:

Calculated for $C_{19}H_{19}NO_4 \cdot H_2O$:

C, 66.46%; H, 6.16%; N, 4.07%.

Found: C, 66.89%; H, 6.46%; N, 3.99%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

3410, 1698, 1634.

Mass Spectrum (m/e):

353 ($M^+ + H_2O$); 325 (M^+).

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 270 MHz), δ ppm:

0.77-0.93 (1H, multiplet),
1.19-1.3 (1H, multiplet),
1.32-1.51 (1H, multiplet),
1.52-1.65 (2H, multiplet),
1.67-1.76 (1H, multiplet),
2.17-2.3 (1H, multiplet),
2.49-2.62 (2H, multiplet),
2.85-3.07 (3H, multiplet),
3.14-3.32 (1H, multiplet),
3.4-3.53 (1H, multiplet),
6,61 (1H, doublet, J=8Hz),
7.0 (1H, triplet, J=8Hz),
7.75 (1H, doublet, J=8Hz).

7-2

Methyl (3aR,12R,12aR,12bS)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-hydroxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylate

7-2(a) Methyl (3aR,12R,12aR,12bS)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-benzyloxy-5-oxoisoquino[2,1,8-lma]-carbazol-12-carboxylate

A suspension consisting of 2.0 g of (3aR,12R,12aR,12bS)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-benzyloxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylic acid and 702 µl of thionyl chloride in 48 ml of methylene chloride was refluxed for 1.5 hour under a nitrogen atmosphere. Subsequently, the solvent was removed by evaporation under reduced pressure. 10 ml of methanol and 38 ml of pyridine were added to the resulting residue and the mixture was then stirred for a further 1.5 hours. After this time, the reaction mixture was poured into 1N aqueous hydrochloric acid and extracted with ethyl acetate. The extract was washed with brine, dried over anhydrous sodium sulfate, and the solvent was removed by evaporation under reduced pressure. The residue was chromatographed on silica gel using a 3 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent, to afford 0.79 g of the title compound.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1718.

Mass Spectrum (m/e):
429 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.86-1.03 (1H, multiplet),

1.13-1.4 (2H, multiplet),
1.49-1.76 (2H, multiplet),
2.17-2.29 (1H, multiplet),
2.48-2.59 (2H, multiplet),
2.62 (1H, doublet of doublets, J=2&17Hz),
2.85-3.0 (3H, multiplet),
3.09-3.17 (1H, multiplet),
3.49 (1H, doublet of doublets, J=2&17Hz),
3.7 (3H, singlet),
5.17 (2H, singlet),
6.77 (1H, doublet, J=8Hz),
7.18 (1H, triplet, J=8Hz),
7.3-7.51 (5H, multiplet),
8.01 (1H, doublet, J=8Hz).

7-2(b) Methyl (3aR,12R,12aR,12bS)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-hydroxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylate

Following a procedure similar to that of Example 6(a), but using 160 mg of the compound obtained in Example 7-2(a), and other agents in appropriate amounts, 83 mg of the title compound was obtained, melting at 255-258°C.

$[\alpha]_D^{23}$ +101.12° (c=0.98, dimethylsulfoxide).

Elemental analysis:
Calculated for $C_{20}H_{21}NO_4$:
C, 70.78%; H, 6.24%; N, 4.13%.
Found: C, 70.70%; H, 6.44%; N, 4.12%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3350, 1731, 1677.

Mass Spectrum (m/e):
339 (M^+).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:

0.61-0.79 (1H, multiplet),
0.95-1.14 (1H, multiplet),
1.22-1.41 (1H, multiplet),
1.48-1.69 (3H, multiplet),
2.1-2.24 (1H, multiplet),
2.4-2.55 (1H, multiplet),
2.76-2.89 (1H, multiplet),
2.91-3.14 (4H, multiplet),
3.28-3.37 (1H, multiplet),
3.64 (3H, singlet),
6,63 (1H, doublet, J=8Hz),
7.0 (1H, triplet, J=8Hz),
7.68 (1H, doublet, J=8Hz),
9.61 (1H, singlet).

7-3 (3aR,12S,12aR,12bS)-12-Amino-2,3,3a,4,11,12,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

7-3(a) Benzyl-N-[(3aR,12S,12aR,12bS)-2,3,3a,4,5,11,12,12a,12b-octahydro-10-benzyloxyisoquino[2,1,8-lma]carbazol-2-yl]carbamate

Following a procedure similar to that of Example 1(e), 0.79 g of the compound obtained in Example 7-2(a) was treated with an alkaline solution to afford 0.734 g of a mixture of epimers at the C-12 position of the carboxylic acid. 400 mg of the mixture of epimers was converted to the carbamate following a procedure similar to that of Example 1(f), using appropriate agents in suitable quantities. 142 mg of the title compound was isolated by recrystallization from toluene as a single isomer.

The properties of the carboxylic acid were as follows:

Melting point: 228-230°C.

$[\alpha]_D^{23}$ +32.44° (c=1.19, dimethylformamide).

Nuclear Magnetic Resonance Spectrum ($CDC\ell_3$, 270 MHz), δ ppm:

0.6-0.8 (1H, multiplet),
0.9-1.15 (1H, multiplet),
1.21-1.67 (4H, multiplet),
2.13-2.31 (1H, multiplet),
2.4-2.51 (2H, multiplet),
2.65-3.0 (2H, multiplet),
3.05-3.36 (3H, multiplet),
5.15-5.3 (2H, doublet, J=10Hz),
6.8-6.91 (1H, multiplet),
7.1-7.2 (1H, multiplet),
7.29-7.55 (5H, multiplet),
7.8-7.85 (1H, multiplet),
12.1-12.5 (1H, broad singlet).

The properties of the carbamate were as follows:

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:

3356, 1715, 1691.

Mass Spectrum (m/e):

520 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDC\ell_3$, 270 MHz), δ ppm:

0.83-1.1 (2H, multiplet),
1.18-1.35 (1H, multiplet),
1.45-1.77 (3H, multiplet),
2.15-2.41 (2H, multiplet),
2.48-2.65 (2H, multiplet),
2.89-2.96 (1H, multiplet),
3.25-3.36 (2H, multiplet),

4.1-4.25 (1H, broad singlet),
4.85-4.92 (1H, multiplet),
5.1-5.2 (4H, multiplet),
6.75 (1H, doublet, J=8Hz),
7.18 (1H, triplet, J=8Hz),
7.27-7.45 (10H, multiplet),
8.0 (1H, doublet, J=8Hz).

7-3(b) (3aR,12S,12aR,12bS)-12-Amino-2,3,3a,4,5,11,12,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-1ma]carbazol-5(1H)-one hydrochloride

Following a procedure similar to that of Example 5(d), a suspension of 142 mg of the compound obtained in Example 7-3(a) in 20 ml of methylene chloride was hydrogenated. Other agents were used in appropriate amounts. The title compound was obtained in a yield of 63 mg, melting at >300°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3291, 1715, 1628.

Mass Spectrum (m/e):
296 (M^+-HCl).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
0.66-0.81 (1H, multiplet),
0.83-1.08 (1H, multiplet),
1.12-1.33 (1H, multiplet),
1.5-1.74 (3H, multiplet),
2.16-2.41 (2H, multiplet),
2.63-2.82 (1H, multiplet),
2.99-3.2 (5H, multiplet),
3.55-3.73 (1H, multiplet),
6.66 (1H, doublet, J=8Hz),
7.05 (1H, triplet, J=8Hz),

7.67 (1H, doublet, J=8 Hz),
8.0-8.5 (1H, broad singlet),
9.7-9.83 (1H, broad singlet).

7-4 4-(Dimethylamino)butyl (3aR,12R,12aR,12bS)-1,2,3,3a,-4,5,11,12,12a,12b-decahydro-10-methoxy-5-oxoisoquino-[2,1,8-lma]carbazol-12-carboxylate hydrochloride

A mixture of 400 mg of (3R,12R,12aR,12bS)-10-benzyloxy-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylic acid and 154 µl of thionyl chloride in 20 ml of methylene chloride was refluxed for 2 hours under a nitrogen atmosphere. After this time, the reaction mixture was cooled in an ice bath, and 308 µl of 4-(dimethylamino) butanol in 8 ml of methylene chloride was added to the ice-cooled mixture. The reaction mixture was stirred overnight at this temperature, and then the mixture was poured into a saturated solution of aqueous sodium hydrogencarbonate and extracted with ethyl acetate. The resulting extract was washed with brine, dried over sodium sulfate, and the solvent removed by evaporation under reduced pressure. The residue was purified by silica gel chromatography, using a 100 : 10 : 5 by volume mixture of methylene chloride, ethanol and aqueous ammonia as the eluent, to give 360 mg of 4-(dimethylamino)butyl (3aR,12R,12aR,12bS)-10-benzyloxy-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylate. This compound was hydrogenated using a similar procedure to that described in Example 5(d) to afford 205 mg of the title compound, melting at 130-132°C.

Elemental analysis:

Calculated for $C_{25}H_{33}N_2ClO_4 \cdot H_2O$:
C, 62.69%; H, 7.36%; N, 5.85%; Cl, 7.40%.
Found: C, 62.15%; H, 7.07%; N, 5.68%; Cl, 7.11%.

Infrared Absorption Spectrum (KBr), ν cm^{-1}:
3400, 3200, 1723, 1700.

Mass Spectrum (m/e):
424 (M^+-HCl).

$[\alpha]_D^{23}$ +83.78° (c=0.74, ethanol).

Nuclear Magnetic Resonance Spectrum (270 MHz, tetradeuterated methanol), δ ppm:
0.78-0.96 (1H, multiplet),
1.15-1.52 (2H, multiplet),
1.54-1.78 (7H, multiplet),
2.18-2.30 (1H, multiplet),
2.52-2.62 (2H, multiplet),
2.71 (6H, singlet),
2.9-3.2 (6H, multiplet),
3.45-3.57 (1H, multiplet)
4.1-4.29 (2H, multiplet),
6.62 (1H, doublet, J=8Hz),
7.01 (1H, triplet, J=8Hz),
7.77 (1H, doublet, J=8Hz).

EXAMPLE 8

(3aR,12R,12aR,12bS)-12-Acetylamino-2,3,3a,4,,11,12,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one

8(a) (3aR,12R,12aR,12bS)-12-Acetylamino-2,3,3a,4,11,12,12a,12b-octahydro-10-acetoxyisoquino[2,1,8-lma]carbazol-5(1H)-one

85 ml of acetic anhydride and a catalytic amount of 4-dimethylaminopyridine were added to a solution of 0.1 g of the compound obtained in Example 1(g) in 1 ml

of pyridine and the mixture stirred overnight. After this time, the reaction mixture was poured into 1N aqueous hydrochloric acid, and extracted with ethyl acetate. The extract was washed with brine, dried over anhydrous sodium sulfate, and the solvent was removed by evaporation under reduced pressure. The resulting residue was chromatographed on silica gel using a 100 : 10 : 1 by volume mixture of methlene chloride, ethanol and aqueous ammonia as eluent, to afford 66 mg of the title compound, melting at 170-172°C.

$[\alpha]_D^{23}$ +82.24° (c=0.76, trichloromethane).

Elemental analysis:

Calculated for $C_{22}H_{24}N_2O_4 \cdot 1/4H_2O$:
C, 68.64%; H, 6.41%; N, 7.28%.
Found: C, 68.89%; H, 6.65%; N, 7.00%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1765, 1708, 1674.

Mass Spectrum (m/e):
380 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
0.86-1.06 (2H, multiplet),
1.2-1.43 (2H, multiplet),
1.6-1.74 (2H, multiplet),
2.0 (3H, singlet),
2.15-2.35 (2H, multiplet),
2.37 (3H, singlet),
2.59-2.74 (2H, multiplet),
2.94-3.18 (3H, multiplet),
4.32-4.38 (1H, multiplet),
5.77 (1H, doublet, J=8Hz),
6.97 (1H, doublet, J=8Hz),

7.29 (1H, triplet, J=8Hz),
8.3 (1H, doublet, J=8Hz).

8(b) (3aR,12R,12aR,12bS)-12-Acetylamino-2,3,3a,4,11,12,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-1ma]carbazol-5(1H)-one

A mixture of 50 mg of the compound obtained in Example 8(a) together with 3 pellets of potassium hydroxide in 2 ml of a 1 : 1 by volume mixture of methanol and water was stirred for one hour. After this time, the reaction mixture was acidified with concentrated aqueous hydrochloric acid. The acidified mixture was then stirred for a further one hour, and then the crystals which precipitated were collected by filtration to give 42 mg of the title compound, melting at >300°C.

$[\alpha]_D^{23}$ +103.33° (c=0.03, dimethylsulfoxide).

Elemental analysis:
Calculated for $C_{20}H_{22}N_2O_3 \cdot 1/10H_2O$:
C, 70.61%; H, 6.58%; N, 8.23%.
Found: C, 70.51%; H, 6.60%; N, 8.26%.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3339, 3183, 1715, 1704, 1662.

Mass Spectrum (m/e):
338 (M^+).

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
0.76-1.0 (2H, multiplet),
1.15-1.33 (1H, multiplet),
1.46-1.65 (3H, multiplet),
1.81 (3H, singlet),

2.04-2.2 (2H, multiplet),
2.47-2.55 (1H, multiplet),
2.75-3.1 (3H, multiplet),
3.19-3.27 (1H, multiplet),
4.02-4.1 (1H, multiplet),
6.72 (1H, doublet, J=8Hz),
7.0 (1H, triplet, J=8Hz),
7.7 (1H, doublet, J=8Hz),
7.97 (1H, doublet, J=8Hz),
9.57 (1H, singlet).

Example 9

(2S,3aS)-2-Amino-1,2,3,3a,4,5-hexahydro-1H-pyrido-[3,2,1-jk]carbazol-6-one hydrochloride

and

(2R,3aR)-2-Amino-1,2,3,3a,4,5-hexahydro-1H-pyrido-[3,2,1-jk]carbazol-6-one hydrochloride

9(a) (1S)-Phenethyl N-[(2S,3aS)-2,3,3a,4,5,6-hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-yl]carbamate

and

(1S)-Phenethyl-N-[(2R,3aR)-2,3,3a,4,5,6-hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazol-2-yl]carbamate

Following a procedure similar to that of Example 3(g), but using a racemic preparation of 354 mg of (2ȝ,3aȝ)-2,3,3a,4,5,6-hexahydro-6-oxo-1H-pyrido[3,2,1-jk]carbazole-2-carboxylic acid and other agents in appropriate amounts, the title compounds were obtained.

The less polar compound had the following properties:

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.4-1.8 (5H, multiplet),
2.0-2.2 (1H, multiplet),
2.3-2.5 (1H, multiplet),
2.7-3.0 (5H, multiplet),
4.4-4.5 (1H, multiplet),
4.95-5.05 (1H, multiplet),
5.75-5.9 (1H, multiplet),
7.25-7.45 (8H, multiplet),
8.39 (1H, doublet, J=8.0Hz).

The more polar compound had the following properties:

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

1.5-1.8 (5H, multiplet),
2.1-2.25 (1H, multiplet),
2.4-2.5 (1H, multiplet),
2.65-3.05 (5H, multiplet),
4.4-4.5 (1H, multiplet),
4.9-5.05 (1H, multiplet),
5.75-5.9 (1H, multiplet),
7.2-7.4 (8H, multiplet),
8.38 (1H, doublet, J=8.0Hz).

9(b) (2S,3aS)-2-Amino-1,2,3,3a,4,5-hexahydro-6H-pyrido-[3,2,1-jk]carbazol-6-one hydrochloride or its enantiomer

The title compound was obtained, following a procedure similar to that of Example 6(g), but using the whole of the less polar isomer of the products in Example 9(a), and other agents in appropriate amounts.

Melting point: 278-285°C.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
1.6-1.8 (2H, multiplet),
2.05-2.15 (1H, multiplet),
2.3-2.4 (1H, multiplet),
2.7-3.1 (5H, multiplet),
3.9-4.0 (1H, multiplet),
7.25-7.55 (3H, multiplet),
8.2-8.4 (4H, multiplet).

$[\alpha]_D$ +85.6° (c=1.00, methanol).

9(c) (2R,3aR)-2-Amino-1,2,3,3a,4,5-hexahydro-6H-pyrido-[3,2,1-jk]carbazol-6-one hydrochloride or its enantiomer

The title compound was obtained, following a procedure similar to that of Example 6(g), but using the whole of the more polar isomer of the products in Example 9(a), and other agents in appropriate amounts.

Melting point: 178-190°C.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 270 MHz), δ ppm:
1.6-1.8 (2H, multiplet),
2.05-2.15 (1H, multiplet),
2.3-2.4 (1H, multiplet),
2.7-3.1 (5H, multiplet),
3.9-4.0 (1H, multiplet),
7.25-7.55 (3H, multiplet),
8.2-8.4 (4H, multiplet).

$[\alpha]_D$ -87.7° (c=1.03, methanol).

Example 10

(3aR,12R,12aR,12bS)-12-Amino-9-benzyloxy-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-1ma]carbazol-5(1H)-one hydrochloride

10(a) Allyl (E)-3-{1-[2-(2-cyclopenten-1-yl)acetyl]-1H-indol-3-yl}-2-propenoate

3.29 g of sodium hydride (55% w/v dispersion in mineral oil) was added to an ice-cooled solution of 24.67 g of allyl 3-[5-benzyloxy-1H-indol-3-yl]-2-propenoate in 160 ml of anhydrous tetrahydrofuran, under a nitrogen atmosphere. The resulting mixture was stirred for 30 minutes, and then a solution of 11.96 g of 2-(2-cyclopenten-1-yl)acetyl chloride in 6 ml of tetrahydrofuran was added to the reaction mixture which was then stirred for a further 30 minutes at 0°C. The reaction mixture was next poured into 800 ml of a dilute aqueous solution of hydrogen chloride and extracted with dichloromethane. The resulting extract was washed with water and brine, dried over anhydrous sodium sulfate and then the solvent was removed by evaporation *in vacuo*. The residue was chromatographed on silica gel using a 1 : 1 by volume mixture of cyclohexane and tetrahydrofuran as eluent. The eluate was crystallized from a mixture of ethyl acetate and cyclohexane to give 27.07 g of the title compound as crystals, melting at 120-121°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1705, 1634.

Mass Spectrum (m/e):
455 (M^+).

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

8.42 (1H, doublet, J=9 Hz),
7.81 (1H, doublet, J=16 Hz),
7.70 (1H, singlet),
7.52-7.78 (6H, multiplet),
7.10 (1H, doublet of doublets, J=3, 9 Hz),
6.50 (1H, doublet, J=16 Hz),
5.95-5.75 (1H, multiplet),
5.64-5.60 (1H, multiplet),
5.43-5.28 (1H, multiplet),
5.15 (2H, singlet),
4.74 (2H, doublet, J=6 Hz),
2.87 (3H, broad singlet),
2.06-1.34 (6H, multiplet).

10(b) (3aR,12R,12aR,12bS)-9-Benzyloxy-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxoisoquino[2,1,8-1ma]carbazol-12-carboxylic acid

The allyl ester of the title compound was obtained by a procedure similar to that of Example 1(c) and 1(d), but using the appropriate types and amounts of starting materials. 0.60 g of triphenylphosphine, 0.60 g of tetrakis(triphenylphosphine)palladium and 17.28 g of sodium 2-ethylhexanoate were then added to a mixture of 23.7 g of the allyl ester of the title compound in 360 ml of ethyl acetate. The reaction mixture was stirred for 25 hours at room temperature and, during this time, 1.36 g of tetrakis(triphenylphosphine)-palladium was added to the reaction mixture in two portions. The reaction mixture was then poured into an aqueous solution of sodium hydrogencarbonate. The aqueous phase was acidified with concentrated aqueous hydrochloric acid and extracted with dichloromethane. The resulting extract was dried over anhydrous sodium sulfate, and the solvent was removed by evaporation

under reduced pressure. The resulting residue was reprecipitated from ethyl acetate to give 10.36 g of the title compound, melting at 247-249°C.

$[\alpha]_D^{25}$ +116.4° (c=0.503, trichloromethane).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
1729, 1669, 1631.

Mass Spectrum (m/e):
415 (M^+).

Elemental analysis:
Calculated for $C_{26}H_{25}NO_4$:
C, 75.16%; H, 6.07%; N, 3.37%.
Found: C, 74.97%; H, 5.98%; N, 3.30%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
8.25 (1H, doublet, J=9 Hz),
7.48-6.95 (7H, multiplet),
5.12 (2H, singlet),
3.19-2.58 (6H, multiplet),
2.19-2.09 (1H, multiplet),
1.69-1.25 (8H, multiplet).

10(c) (3aR,12R,12aR,12bS)-12-Amino-9-benzyloxy-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

Following a procedure similar to that of Example 1(f), but using the appropriate types and amounts of starting materials, 4.86 g of 4-methoxybenzyl N-(3aR,12R,12aR,12bS)-9-benzyloxy-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxoisoquino[2,1,8-lma]carbazol-12-yl]-carbamate was obtained. A solution of 4N HCl in 6.5 ml of dioxane was added to a solution of 4.78 g of the

carbamate in 96 ml of dioxane. The resulting mixture was heated to 80°C and maintained at this temperature for 3 hours. After this time, the solvent was removed by evaporation in vacuo. The resulting residue was recrystallized from a mixture of dichloromethane and isopropyl ether to give 3.11 g of the title compound as crystals, melting at 176-179°C.

$[\alpha]_D^{25}$ +68.3° (c=0.503, methanol).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3400, 1702.

Mass Spectrum (m/e):
386 (M^+).

Elemental analysis:
Calculated for $C_{25}H_{27}ClN_2O_2$:
C, 71.16%; H, 6.21%; N, 6.64%; Cl, 8.40%.
Found: C, 70.85%; H, 6.51%; N, 6.58%; Cl, 8.18%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:
8.33 (2H, broad singlet),
8.24 (1H, singlet),
7.43-6.90 (7H, multiplet),
5.05 (2H, doublet of doublets, J=11,15Hz),
3.47 (1H, broad singlet),
3.38 (1H, broad singlet),
3.01 (1H, doublet, J=17Hz),
2.83 (1H, doublet, J=17Hz),
2.68 (1H, doublet of doublets, J=5,17Hz),
2.43 (1H, doublet, J=16Hz),
2.25-2.21 (1H, multiplet),
1.80-1.12 (5H, multiplet),
0.82-0.77 (2H, multiplet).

10(d) (3aR,12R,12aR,12bS)-9-Benzyloxy-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxo-N-[(S)-1-phenethyl]isoquino-[2,1,8-lma]carbazol-12-carboxamide

A drop of dimethylformamide and 0.11 ml of thionyl chloride were both added to a suspension of 0.125 g of the compound obtained in Example 10(b) in 2.5 ml of dichloromethane. The reaction mixture was refluxed for 1 hour, and then the solvent was removed by evaporation under reduced pressure. The resulting residue was redissolved in 2.5 ml of dichloromethane, and then 0.05 ml of (S)-1-phenethylamine and 0.05 ml of pyridine were added to the solution thus obtained, and the resulting mixture was stirred at room temperature for 15 minutes. After this time, the reaction mixture was diluted with ethyl acetate, washed successively with dilute aqueous hydrochloric acid, water, a saturated aqueous solution of sodium hydrogencarbonate and brine, dried on anhydrous sodium sulfate and then the solvent was removed by evaporation under reduced pressure. The residue was purified by silica gel column chromatography using a 1 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent. The eluate was crystallized from a mixture of tetrahydrofuran and hexane to give 0.466 g of the title compound, melting at 249-251°C.

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3360, 1688, 1666, 1639.

Mass Spectrum (m/e):
518 (M^+).

Elemental analysis:
Calculated for $C_{34}H_{34}N_2O_3$:
C, 78.74%; H, 6.61%; N, 5.40%.
Found: C, 79.39%; H, 6.76%; N, 5.19%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 270 MHz), δ ppm:

8.30 (1H, doublet, J=9Hz),
7.49-6.98 (12H, multiplet),
5.94-5.90 (2H, multiplet),
5.12 (2H, singlet),
2.91-2.01 (6H, multiplet),
1.40 (3H, doublet, J=7Hz),
1.68-1.03 (9H, multiplet).

EXAMPLE 11

(3aR,12R,12aR,12bS)-12-Amino-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride

and

(3aS,12S,12aS,12bR)-12-Amino-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride.

11(a) (S)-1-Phenethyl N-[(3aR,12R,12aR,12bS)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxoisoquino[2,1,8-lma]carbazol-12-yl]carbamate

and

(S)-1-Phenethyl N-[(3aS,12S,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-5-oxoisoquino[2,1,8-lma]carbazol-12-yl]-carbamate

A solution of 0.65 g of (3aβ,12β,12aβ,12bβ)-12-amino-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]carbazol-5(1H)-one and 864 mg of (S)-1-phenethyloxycarbonylimidazole in 16 ml of toluene was heated at 100°C for 3 days and then the solvent was removed by evaporation under reduced pressure. The resulting

residue was purified by Lobar chromatography using a 5 : 1 by volume mixture of cyclohexane and ethyl acetate as the eluent to give the title compounds.

The less polar compound was obtained in a yield of 83 mg.

The more polar compound was obtained in a yield of 93 mg.

11(b) (3aR,12R,12aR,12bS)-12-Amino-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride or its enantiomer

Following a procedure similar to that of Example 6(g), but using the appropriate types and amounts of starting materials, the title compound was obtained from the whole of the less polar compound obtained in Example 11(a), melting at 201-202°C.

$[\alpha]_D$ +78.8° (c=1.01, methanol).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3404, 1703, 1684, 1632.

Mass Spectrum (m/e):
316 (M^+).

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 270 MHz), δ ppm:
0.8-1.8 (6H, multiplet),
2.3-2.5 (2H, multiplet),
2.67 (1H, doublet of doublets, J=3,17Hz),
2.85 (1H, doublet, J=18Hz),
3.1-3.4 (3H, multiplet),
3.80-3.85 (1H, multiplet),
7.3-7.5 (3H, multiplet),
8.29-8.23 (1H, multiplet).

11(c) (3aS,12S,12aS,12bR)-12-Amino-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]carbazol-5(1H)-one hydrochloride or its enantiomer

Following a procedure similar to that of Example 6(g), but using the appropriate types and amounts of starting materials, the title compound was obtained from the whole of the more polar compound obtained in Example 11(a), melting at 183-186°C.

$[\alpha]_D$ -46.3° (c=0.57, methanol).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3352, 1704, 1682, 1632.

Mass Spectrum (m/e):
316 (M^+).

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 270 MHz), δ ppm:
0.8-1.8 (6H, multiplet),
2.3-2.5 (2H, multiplet),
2.67 (1H, doublet of doublets, J=3,17Hz),
2.85 (1H, doublet, J=18Hz),
3.1-3.4 (3H, multiplet),
3.80-3.85 (1H, multiplet),
7.3-7.5 (3H, multiplet),
8.29-8.23 (1H, multiplet).

EXAMPLE 12

12-1 (3aS,12S,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-Decahydro-10-hydroxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylic acid

1 ml of 1M solution of boron tribromide in methylene

chloride was added to a suspension of 500 mg of (3aS,12S,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-methoxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxylic acid in 5 ml of methylene chloride at 0°C, and the resulting mixture was stirred overnight. After this time, 5 ml of water and 5 ml of saturated aqueous sodium hydrogencarbonate were added to the reaction mixture at room temperature. The reaction mixture was then stirred for 30 minutes, and then the aqueous layer was washed with methylene chloride and acidified with concentrated aqueous hydrochloric acid. The precipitate was collected to afford 343 mg of the title compound.

Infrared Absorption Spectrum (KBr), ν cm^{-1}:
3402, 1705.

Mass Spectrum (m/e):
325 (M^+).

Nuclear Magnetic Resonance Spectrum (270 MHz, tetradeuterated methanol), δ ppm:
0.79-0.96 (1H, multiplet),
1.15-1.52 (2H, multiplet),
1.53-1.79 (3H, multiplet),
2.18-2.3 (1H, multiplet),
2.5-2.6 (2H, multiplet),
2.85-3.07 (3H, multiplet,
3.13-3.22 (1H, multiplet),
3.41-3.53 (1H, multiplet),
6.61 (1H, doublet, J=8Hz),
7.00 (1H, triplet, J=8Hz),
7.74 (1H, doublet, J=8Hz).

12-2 Methyl (3aS,12S,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,-12b-decahydro-10-hydroxy-5-oxoisoquino[2,1,8-1ma]-carbazol-12-carboxylate

A solution of 100 mg of the compound obtained in Example 12-1 in 20 ml of methanol was treated with an excess of a solution of diazomethane in diethyl ether at 0°C. The reaction mixture was dried by evaporation under reduced pressure and the residue was recrystallized from a mixture of hexane and acetone to afford 62 mg of the title compound melting at 253-256°C.

$[\alpha]_D^{23}$ -90.0° (c=0.63, dimethylsulfoxide).

Elemental analysis:

Calculated for $C_{20}H_{21}NO_4$ $1/3H_2O$:

C, 69.55%; H, 6.32%; N, 4.06%.

Found: C, 69.72%; H, 6.35%; N, 4.31%.

Infrared Absorption Spectrum (KBr), ν cm^{-1}:

3360, 1732, 1676.

Mass Spectrum (m/e):

339 (M^+).

Nuclear Magnetic Resonance Spectrum (270 MHz, hexadeuterated dimethylsulfoxide), δ ppm:

0.82-0.99 (1H, multiplet),
1.12-1.45 (2H, multiplet),
1.5-1.8 (3H, multiplet),
2.19-2.3 (1H, multiplet),
2.5-2.7 (2H, multiplet),
2.85-3.2 (4H, multiplet),
3.41-3.53 (1H, multiplet),
3.71 (3H, singlet),
6.69 (1H, doublet, J=8Hz),
7.01 (1H, triplet, J=8Hz),

7.81 (1H, doublet, J=8Hz),
9.12 (1H, singlet).

12-3 (3aS,12S,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-Decahydro-10-hydroxy-5-oxoisoquino[2,1,8-lma]carbazol-12-carboxamide

A mixture of 500 mg of (3aS,12S,12aS,12bR)-1,2,3,-3a,4,5,11,12,12a,12b-decahydro-10-methoxy-5-oxoisoquino-[2,1,8-lma]carbazol-12-carboxylic acid, 210µl of chloroethylformate, and 247µl of triethylamine in 6.7 ml of acetone was stirred for 30 minutes at 0°C. After this time, 1.7 ml of aqueous ammonia was added to the mixture and which was then stirred for a further one hour. At the end of this time, water was added to the reaction mixture which was then extracted with ethyl acetate. The extract was dried over anhydrous sodium sulfate and dried by evaporation under reduced pressure. The residue was recrystallized from a mixture of hexane and acetone to give 356 mg of (3aS,12S,12aS,12bR)-1,2,3,3a,4,5,11,12,12a,12b-decahydro-10-methoxy-5-oxoiso-quino[2,1,8-lma]carbazol-12-carboxamide. 1 ml of a 1M solution of boron tribromide in methylene chloride was added to a suspension of 300 mg of this compound in 3 ml of methylene chloride at 0°C, and the resulting mixture was stirred overnight at room temperature. 1 ml of water and 1 ml of a saturated aqueous sodium hydrogencarbonate solution were then added to the mixture which was then stirred for a further 30 minutes. At the end of this time, a large excess of acetone was added to the reaction mixture, and the resulting mixture was dried over anhydrous sodium sulfate and the solvent removed by evaporation under reduced pressure. The residue was recrystallised from a mixture of hexane and acetone to afford 171 mg of the title compound, melting at 194-195°C.

$[\alpha]_D^{23}$ -66.59° (c=0.85, dimethylsulfoxide).

Infrared Absorption Spectrum (KBr), ν cm^{-1}:
3343, 3214, 1702,1668.

Mass Spectrum (m/e):
324 (M^+).

Nuclear Magnetic Resonance Spectrum (270 MHz, hexadeuterated acetone), δ ppm:
0.77-0.93 (1H, multiplet),
1.1-1.45 (2H, multiplet),
1.5-1.85 (3H, multiplet),
2.18-2.31 (1H, multiplet),
2.4-2.57 (2H, multiplet),
2.76-2.83 (3H, multiplet),
2.85-3.08 (2H, multiplet),
3.2-3.37 (2H, multiplet),
6.64 (1H, doublet, J=8Hz),
7.0 (1H, triplet, J=8Hz),
7.81 (1H, doublet, J=8Hz),
8.46 (1H, singlet).

EXAMPLE 13

(3aS,12S,12aS,12bR)-12-Acetylamino-2,3,3a,4,11,12,12a,-12b-octahydro-10-hydroxyisoquino[2,1,8-lma]carbazol-5(1H)-one

13(a) (3aS,12S,12aS,12bR)-12-Acetylamino-10-acetoxy-2,3,3a,4,11,12,12a,12b-octahydroisoquino[2,1,8-lma]-carbazol-5(1H)-one

Following a procedure similar to that of Example 8(a), but starting with 360 mg of (3aS,12S,12aS,12bR)-12-amino-2,3,3a,4,11,12,12a,12b-octahydro-10-hydroxy-

isoquino[2,1,8-lma]carbazol-5(1H)-one (as obtained in Example 11c) and using 284µl of acetic anhydride, 5 ml of pyridine and a catalytic amount of dimethylaminopyridine, 344 mg of the title compound were obtained, melting at 208-210°C.

Mass Spectrum (m/e):
380 (M^+).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3280, 1760, 1735, 1703.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$), δ ppm:
0.8-1.8 (6H, multiplet),
2.04 (3H, singlet),
2.1-2.4 (2H, multiplet),
2.37 (3H, singlet),
2.6-2.8 (2H, multiplet),
2.9-3.2 (3H, multiplet),
4.3-4.4 (1H, multiplet),
6.99 (1H, doublet J=8Hz),
7.29 (1H, triplet, J=8Hz),
8.30 (1H, doublet, J=8Hz).

13(b) (3aS,12S,12aS,12bR)-12-Acetylamino-2,3,3a,4,11,-12,12a,12b-octahydro-10-hydroxyisoquino[2,1,8-lma]-carbazol-5(1H)-one

Following a procedure similar to that of Example 8(b), starting with 204 mg of the product of Example 13(a) and using 3 ml of a 1N aqueous sodium hydroxide solution and 5 ml of ethanol, 110 mg of the title compound was obtained, melting at >300°C.

Mass Spectrum (m/e):
338 (M^+), 279 (M^+-acetic acid).

Infrared Absorption Spectrum (KBr), ν_{max} cm^{-1}:
3330, 3170, 1710, 1700, 1660.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide), δ ppm:
0.6-1.7 (6H, multiplet),
1.81 (3H, singlet),
2.0-2.2 (2H, multiplet),
2.5-2.6 (1H, multiplet),
2.79 (1H, doublet, J=18Hz),
2.9-3.1 (2H, multiplet),
3.22 (1H, broad singlet)
4.0-4.1 (1H, multiplet),
6.62 (1H, doublet J=8Hz),
7.00 (1H, triplet, J=8Hz),
7.70 (1H, doublet, J=8Hz),
7.98 (1H, doublet, J=7Hz),
9.60 (1H, singlet).

M&C FOLIO:230P70859/FP-9414 WANGDOC: 1012D

The compound numbers used in the following Examples correspond to those in the accompanying Reaction Schemes.

PREPARATION OF 3-TETRAHYDROPYRIDINYL-1H-INDOLES

EXAMPLE 14

3-(1-Benzyl-1,2,5,6-tetrahydropyridin-4-yl)-1H-indole Compound (2a)

30.0 g of indole (0.256 mol) were dissolved in 500 ml of methanol containing 83.0 g of sodium methoxide (1.54 mol, 6 equivalents). 72.7 g of 1-benzyl-4-piperidinone (0.384 mol, 1.5 equivalents) were added to the reaction mixture and the resulting solution was refluxed under an argon atmosphere for 8 hours. After this time, the reaction mixture was cooled and the methanol was evaporated to half its original volume under reduced pressure. A white crystalline precipitate was obtained, and this was filtered off, washed successively with several portions of methanol and dried by evaporation *in vacuo* to give 53.2 g of the title compound in a yield of 72%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

2.60 (2H, broad singlet),
2.78 (2H, triplet, J=5.7Hz),
3.27 (2H, doublet of triplets, J=2.8 and 3.2Hz),
3.68 (2H, singlet),
6.20 (1H, multiplet),
7.13 (1H, triplet, J=7Hz),
7.15 (1H, doublet, J=2.7Hz),

7.19 (1H, triplet, J=7Hz),
7.28 (1H, doublet, J=7Hz),
7.33-7.36 (3H, multiplet),
7.40 (2H, doublet, J=8Hz),
7.88 (1H, doublet, J=8Hz),
8.16 (1H, broad singlet).

EXAMPLE 15

3-(1-Benzyl-1,2,5,6-tetrahydropyridin-3-yl)-1H-indole Compound (1a)

The title compound was obtained following a procedure similar to that of Example 14, but starting with 15.6 g of indole and 20.0 g of 1-benzyl-3-piperidone. The resulting residue was chromatographed on silica gel using, as eluent, a 1 : 1 by volume mixture of hexane and ethyl acetate. The eluate was then recrystallised from the same solvent to give 11.9 g of straw-coloured crystalline solid in a yield of 31%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:
2.40 (2H, multiplet),
2.68 (2H, triplet, J=5.8Hz),
3.38 (2H, doublet, J=2Hz),
3.71 (2H, singlet),
6.25 (1H, multiplet),
7.03 (1H, doublet, J=2.5Hz),
7.13 (1H, triplet, J=8Hz),
7.18 (1H, triplet, J=8Hz),
7.27 (1H, doublet, J=7Hz),
7.31-7.42 (5H, multiplet),
7.85 (1H, doublet, J=8Hz),
8.18 (1H, broad singlet).

EXAMPLE 16

5-Methoxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)-1H-indole
Compound (1b)

The title compound was obtained following a procedure similar to that of Example 14, but starting with 8.5 g of 5-methoxyindole and 18.4 g of 1-benzyl-3-piperidone. The resulting residue was chromatographed on silica gel using, as eluent, a 1 : 1 by volume mixture of hexane and ethyl acetate to give 8.5 g of the product as a straw-coloured powder in a yield of 49%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

2.41 (2H, multiplet),
2.69 (2H, triplet, J=5.7Hz),
3.37 (2H, doublet, J=2Hz),
3.71 (2H, singlet),
3.84 (3H, singlet),
6.18 (1H, multiplet),
6.85 (1H, doublet of doublets, J=8.8 and 2.4Hz),
7.03 (1H, doublet, J=2.4Hz),
7.21-7.42 (7H, multiplet),
8.06 (1H, broad singlet).

EXAMPLE 17

5-Benzyloxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)-1H-indole
Compound (1c)

The title compound was obtained following a procedure similar to that of Example 14, but starting with 3.5 g of 5-benzyloxyindole and 8.9 g of

1-benzyl-3-piperidinone, 4.5 g of the title compound was obtained in a yield of 73%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

2.40 (2H, multiplet),
2.69 (2H, triplet, J=5.7Hz),
3.36 (2H, broad singlet),
3.71 (2H, singlet),
5.08 (2H, singlet),
6.14 (1H, multiplet),
6.93 (1H, doublet of doublets, J=8.8 and 2.4Hz),
7.04 (1H, doublet, J=2.4Hz),
7.17-7.51 (12H, multiplet),
8.02 (1H, broad singlet).

EXAMPLE 18

5-Methoxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)-1H-indole
Compound (2b)

Following a procedure similar to that of Example 14, but starting with 15 g of 5-methoxyindole and 55 g of 1-benzyl-4-piperidinone, 25 g of the title compound was obtained in a yield of 78%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

2.59 (2H, multiplet),
2.75 (2H, triplet, J=5.7Hz),
3.36 (2H, doublet of triplets, J=3.2 and 2.5Hz),
3.67 (2H, singlet),
3.84 (3H, singlet),
6.12 (1H, multiplet),
6.85 (1H, doublet of doublets, J=8.8 and 2.4Hz),

7.12 (1H, doublet, J=2.6Hz),
7.23-7.42 (7H, multiplet),
8.01 (1H, broad singlet).

EXAMPLE 19

5-Benzyloxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)-1H-indole
Compound (2c)

The title compound was prepared following a procedure similar to that of Example 14, but starting with 12 g of 5-benzyloxyindole and 36 g of 1-benzyl-4-piperidinone. The crude product was chromatographed on a silica gel column using, as eluent, a 1 : 2 by volume mixture of ethyl acetate and toluene to afford 15 g of the title compound as a white powder in a yield of 71%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:
2.58 (2H, multiplet),
2.75 (2H, triplet, J=5.7Hz),
3.24 (2H, doublet of triplets, J=3.2 and 2.5Hz),
3.67 (2H, singlet),
5.09 (2H, singlet),
6.08 (1H, multiplet),
6.93 (1H, doublet of doublets, J=8.8 and 2.4Hz),
7.12 (1H, doublet, J=2.6Hz),
7.23-7.48 (12H, multiplet),
7.99 (1H, broad singlet).

[EXAMPLE 20]

EXAMPLE 21

3-(1-t-Butoxycarbonyl-1,4,5,6-tetrahydropyridin-3-yl)-1H-indole
Compound (11)

Following a procedure similar to that of Example 14, but starting with 6.0 g of indole and 15.3 g 1-t-butoxycarbonyl-3-piperidinone, the title compound was obtained by chromatography of the reaction mixture on a silica gel column at 40 - 60°C using, as eluent, a 1 : 1 by volume mixture of ethyl acetate and petroleum ether. The title compound was afforded as a colourless oil which later solidified. The yield was 59%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

1.52 (9H, singlet),
1.99 (2H, multiplet),
2.47 (2H, broad singlet),
3.67 (2H, broad singlet),
7.12-7.23 (3H, multiplet),
7.33-7.36 (2H, multiplet),
7.84 (1H, doublet, J=7.7Hz),
8.19 (1H, multiplet).

ACYLATION OF 3-CYCLOALKENYL INDOLES

EXAMPLE 22

1-(4-Pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride
Compound (7a)

A suspension of 0.032 mol of sodium hydride (a 60% w/v dispersion in mineral oil) in 30 ml of anhydrous tetrahydrofuran (a 60% w/v mixture) was added to a cold solution of 8.50 g (0.030 mol) of 3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)-1H-indole (compound 2a, as obtained in Example 14) in 300 ml of dry tetrahydrofuran. The mixture was stirred at room temperature under an argon atmosphere for 1 hour and then cooled to a temperature of 0 - 5°C. A solution of 3.67 g (0.031 mol) of 4-pentenoyl chloride in 70 ml of tetrahydrofuran was then added to the resulting mixture which was then stirred for a further 2 hours. The reaction mixture was then acidified with 2N aqueous hydrochloric acid and the resulting white precipitate was collected by filtration and washed successively with tetrahydrofuran, water and diethyl ether and then dried by evaporation in vacuo. 8.75 g of the title compound was obtained in a yield of 73%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$/ tetradeuterated methanol, 400 MHz), δ ppm:

2.45 (2H, multiplet),
2.55 (2H, broad peak),
2.94 (2H, triplet, J=7.4Hz),
3.70 (2H, broad singlet),
3.90 (2H, singlet),
4.18-4.35 (2H, multiplet),
4.94 (1H, doublet of doublets, J=1.2 and 10.2Hz),

5.03 (1H, doublet of doublets, J=1.6 and 17.1Hz),
5.77-5.86 (1H, multiplet),
6.08 (1H, multiplet),
7.19 (1H, triplet, J=8Hz),
7.22-7.29 (1H, multiplet),
7.32-7.38 (2H, multiplet),
7.48 (1H, singlet),
7.51 (2H, multiplet),
7.61 (1H, doublet, J=8Hz),
8.36 (1H, doublet, J=8Hz).

EXAMPLE 23

1-(4-Pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride
Compound (3a)

Following a procedure similar to that of Example 22, but using 5.32 g of 3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)-1H-indole (compound 1a, as obtained in Example 15), the title compound was obtained in a yield of 72%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, tetradeuterated methanol, 400 MHz), δ ppm:
2.59 (2H, multiplet),
2.71 (2H, broad singlet),
3.07 (2H, triplet, J=7.3Hz),
3.23 (2H, broad singlet),
3.93 (2H, broad singlet),
4.28 (2H, broad singlet),
5.08 (1H, doublet of doublets, J=1.3 and 10.2Hz),
5.17 (1H, doublet of doublets, J=1.7 and 17.1Hz),
5.95 (1H, multiplet),
6.43 (1H, multiplet),
7.31 (1H, triplet, J=7Hz),

7.39 (1H, triplet, J=7Hz),
7.47 (3H, multiplet),
7.60 (3H, multiplet),
7.72 (1H, doublet, J=8Hz),
8.48 (1H, doublet, J=8Hz).

EXAMPLE 24

5-Methoxy-1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride Compound (7b)

Following a procedure similar to that of Example 22, a reaction mixture obtained from 8.5 g of 5-methoxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)-1H-indole (compound 2b, as obtained in Example 18) and 7.9 g of 4-pentenoyl chloride was partitioned between ethyl acetate and dilute ammonia. The organic layer was removed, washed with water and taken to dryness by evaporation in vacuo. The resulting residue was chromatographed on a silica gel column using, as eluent, a 3 : 7 by volume mixture of ethyl acetate and petroleum ether (boiling point 60-80°C), to give the product as a light yellow oil. This was dried and then dissolved in ethylacetate and then treated with a solution of hydrogen chloride in ether. The resulting light brown precipitate was collected by filtration, washed with ether and dried in vacuo at 50°C for four hours to afford the title compound in a yield of 87%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz, 365°K), δ ppm:

2.45-2.61 (2H, multiplet),
2.90 (2H, broad singlet),
3.10 (2H, triplet, J=7Hz),
3.52 (2H, broad singlet),

3.82 (3H, singlet),
3.93 (2H, broad singlet),
4.45 (2H, broad singlet),
5.11-5.38 (2H, multiplet),
5.93 (1H, multiplet),
6.25 (1H, broad singlet),
7.05 (1H, doublet of doublets, J=2.3 and 9Hz),
7.32 (1H, doublet, J=2.3Hz),
7.50-7.68 (5H, multiplet),
7.83 (1H, broad singlet),
8.32 (1H, doublet, J=9Hz).

EXAMPLE 25

1-(4-Pentenoyl)-3-(1-t-butoxycarbonyl-1,4,5,6-tetrahydropyridin-3-yl)indole
Compound (12)

Following a procedure similar to that of Example 22, a reaction mixture obtained from 5.2 g of 3-(1-t-butoxycarbonyl-1,4,5,6-tetrahydropyridin-3-yl)-1H-indole (compound 11, as obtained in Example 21) and 5.2 g of 4-pentenoyl chloride in 150 ml of tetrahydrofuran was taken to dryness by evaporation *in vacuo*. 10 ml of tetrahydrofuran containing 1 equivalent of acetic acid was added to the reaction mixture, and then water was added, after which the reaction mixture was extracted with diethyl ether. The diethyl ether was removed by evaporation under reduced pressure, and the residue was stirred into a small volume of hexane and ethyl acetate (4 : 1 v/v), filtered and washed with hexane. The product was obtained in a yield of 69% as a white solid.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:
1.55 (9H, singlet),

2.01 (2H, broad singlet),
2.46 (2H, broad singlet),
2.60 (2H, doublet of triplets, J=6.9 and 7Hz),
3.10 (2H, triplet, J=7Hz),
3.67 (2H, multiplet),
5.06 (1H, doublet, J=10.1 and 1.3Hz),
5.15 (1H, doublet, J=17.1 and 1.3Hz),
5.95 (1H, multiplet),
7.26 and 7.39 (3H, multiplet),
7.50 and 7.65 (1H, two singlets, rotameric mixture),
7.77-7.87 (1H, two doublets, J=7.5Hz, rotameric mixture),
8.53 (1H, multiplet).

PREPARATION OF 1-(4-PENTYNOYL)-3-(1-BENZYL-1,2,5,6-TETRAHYDROPYRIDINYL) INDOLES

EXAMPLE 26

1-(4-Pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride
Compound (16a)

0.36 g (0.015 mol) of sodium hydride (a 60% w/v dispersion in mineral oil) was added to a cold solution of 2.88g of 3-(1-benzyl-1,2,3,6-tetrahydropyridin-3-yl)-1H-indole (compound 1a, obtained as in Example 15) in 130 ml of dry tetrahydrofuran. The mixture was stirred at room temperature under an argon atmosphere for 30 minutes and then cooled to a temperature of 0 - 5°C. A solution of 1.75 g of 4-pentynoyl chloride in 5 ml of tetrahydrofuran was added to the reaction mixture, which was then stirred for a further 2 hours. After this time, the reaction mixture was acidified with 2N aqueous

hydrochloric acid and the resulting white precipitate was collected by filtration, washed successively with tetrahydrofuran, water and diethyl ether and then dried in vacuo to give 3.08 g of the title compound in a yield of 76%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.60-2.65 (4H, multiplet),
2.69 (1H, triplet, J=2.5Hz),
3.26 (2H, triplet, J=6.7Hz),
3.30 (2H, multiplet),
4.15 (2H, singlet),
4.42 (2H, singlet),
6.53 (1H, multiplet),
7.39 (1H, triplet, J=8Hz),
7.45 (1H, triplet, J=8Hz),
7.50-7.57 (5H, multiplet),
7.87 (1H, doublet, J=7Hz),
7.88 (1H, singlet),
8.43 (1H, doublet, J=8Hz).

EXAMPLE 27

5-Methoxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride Compound (16b)

Following a procedure similar to that of Example 26, but starting with 1.59 g of 5-methoxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole (compound 1b, as obtained in Example 16), 1.69 g of the title compound was obtained in a yield of 78%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.57 (1H, triplet, J=2.4Hz),
2.60-2.66 (4H, multiplet),
3.22 (2H, triplet, J=6.4Hz),
3.4-3.7 (2H, broad peak),
3.82 (3H, singlet),
4.08 (2H, singlet),
4.48 (2H, singlet),
6.49 (1H, multiplet),
7.06 (1H, doublet of doublets, J=9.1 and 2.2Hz),
7.24 (1H, doublet, J=2.2Hz),
7.36-7.60 (5H, multiplet),
7.77 (1H, singlet),
8.32 (1H, doublet, J=9.1Hz).

EXAMPLE 28

5-Benzyloxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride Compound (16c)

Following a procedure similar to that of Example 26, but starting with 4.0 g of 5-benzyloxy-3-(1-benzyl-1,2,5,6-tetra- hydropyridin-3-yl)indole (compound 1c, as obtained in Example 17), 3.41 g of the title compound was obtained in a yield of 65%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:
2.59-2.65 (5H, multiplet),
3.20 (2H, triplet, J=6.5Hz),
3.40 (2H, broad singlet),
4.05 (2H, singlet),
4.45 (2H, singlet),
5.16 (2H, singlet),
6.40 (1H, multiplet),
7.11 (1H, doublet of doublets, J=9.0 and 2.2Hz),

7.28 (1H, doublet, J=2.2Hz),
7.36-7.58 (10H, multiplet),
7.77 (1H, singlet),
8.32 (1H, doublet, J=9.0Hz).

EXAMPLE 29

1-(4-Pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride
Compound (19a)

Following a procedure similar to that of Example 26, but starting with 9.0 g of 3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole (compound 2a, as obtained in Example 14), 9.53 g of the title compound was obtained in a yield of 76%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:
2.61 (2H, triplet of doublets, J=6.8 and 2.4Hz),
2.72 (1H, triplet, J=2.4Hz),
2.87 (2H, broad singlet),
3.29 (2H, triplet, J=6.8Hz),
3.45 (2H, broad singlet),
3.84 (2H, singlet),
4.39 (2H, singlet),
6.35 (1H, broad singlet),
7.37 (1H, triplet, J=8Hz),
7.43 (1H, triplet, J=8Hz),
7.51-7.57 (5H, multiplet),
7.91 (1H, doublet, J=8Hz),
7.96 (1H, singlet),
8.44 (1H, doublet, J=8Hz).

EXAMPLE 30

5-Methoxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride Compound (19b)

Following a procedure similar to that of Example 26, but starting with 3.18 g 5-methoxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole (compound 2b, as obtained in Example 18), 3.77 g of the title compound was obtained in a yield of 89%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.61 (2H, triplet of doublets, J=6.7 and 2.4Hz),
2.68 (1H, triplet, J=2.4Hz),
2.86 (2H, broad singlet),
3.25 (2H, triplet, J=6.7Hz),
3.47 (2H, broad singlet),
3.82 (3H, singlet),
3.84 (2H, singlet),
4.39 (2H, singlet),
6.31 (1H, broad singlet),
7.04 (1H, doublet of doublets, J=9.0 and 2.3Hz),
7.32 (1H, doublet, J=2.3Hz),
7.52-7.57 (5H, multiplet),
7.91 (1H, singlet),
8.34 (1H, doublet, J=9.0Hz).

EXAMPLE 31

5-Benzyloxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride Compound (19c)

Following a procedure similar to that of Example 26,

but starting with 3.95 g of 5-benzyloxy-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole (compound 2c, as obtained in Example 19), 3.89 g of the title compound was obtained in a yield of 76%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.61 (2H, triplet of doublets, J=6.8 and 2.4Hz),
2.68 (1H, triplet, J=2.4Hz),
2.83 (2H, broad singlet),
3.24 (2H, triplet, J=6.8Hz),
3.44 (2H, broad singlet),
3.81 (2H, singlet),
4.36 (2H, singlet),
5.17 (2H, singlet),
6.25 (1H, broad singlet),
7.11 (1H, doublet of doublets, J=9.1 and 2.2Hz),
7.34-7.58 (11H, multiplet),
7.90 (1H, singlet),
8.33 (1H, doublet, J=9.1Hz).

[EXAMPLE 32]

PREPARATION OF PENTACYCLIC TETRAHYDROCARBAZOLES

EXAMPLE 33

3-Benzyl-1,2,3,4,4a,4b,5,5a,6,7-decahydropyrido[3,4-c]-pyrido[1,2,3-lm]carbazol-8-one
Compound (9a)

Method 1

5.55 g (0.014mol) of 1-(4-pentenoyl)-3-(1-benzyl-

1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride (compound 7a, as obtained in Example 22) were suspended in 550 ml of diphenyl ether and gradually heated to 230°C under a slow stream of argon. The reaction mixture was stirred at 230°C for up to 4 hours until the hydrochloride starting material was fully consumed. The solvent was then removed in vacuo and the residue chromatographed on a silica gel column using ethyl acetate as eluent. 3.75 g of the crude product (yield of 74%) was obtained after recrystallization from ethyl acetate as light green needles.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz, 375K), δ ppm:

1.45-1.65 (2H, multiplet),
1.70-1.81 (1H, multiplet),
1.95-2.11 (3H, multiplet),
2.21-2.33 (2H, multiplet),
2.43-2.52 (2H, multiplet),
2.56 (1H, multiplet),
2.63-2.73 (2H, multiplet),
2.81-3.0 (2H, multiplet),
3.40 (2H, singlet),
7.18-7.33 (7H, multiplet),
7.60 (1H, doublet),
8.30 (1H, doublet).

Method 2

0.5 g (0.0013 mol) of 3-benzyl-1,2,3,4,4a,5,5a,6,7,-13c-decahydropyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one (compound 8a, as obtained in subsequent Example 37) was suspended in 70 ml of mesitylene containing a catalytic amount of hydrogen chloride. The reaction mixture was then gradually heated to 150 - 160°C under a slow stream of argon gas for one hour. The solvent was removed by evaporation in vacuo and the residue was recrystallized

from isopropanol to give the title compound in a yield of 83%.

EXAMPLE 34

2-Benzyl-1,2,3,4,4a,4b,5,5a,6,7-decahydropyrido[4,3-c]-pyrido[1,2,3-lm]carbazol-8-one Compound (5a)

Following a procedure similar to that of Example 33, Method 2, but starting from 0.1 g of 2-benzyl-1,2,3,4,-4a,5,5a,6,7,13c-decahydropyrido[4,3-c]pyrido[1,2,3-lm,]-carbazol-8-one (compound 4a, as obtained in Example 36). The title compound was obtained in a yield of 92%.

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 500 MHz, 300K), δ ppm:

1.72 (2H, multiplet),
1.92 (1H, multiplet),
2.16 (3H, multiplet),
2.50 (1H, multiplet),
2.80-2.96 (3H, multiplet),
3.08 (1H, multiplet),
3.27 (1H, multiplet),
3.38 (1H, multiplet),
3.54 (2H, multiplet),
4.20 (1H, doublet, J=13Hz),
4.44 (1H, doublet, J=13Hz),
7.01 (1H, multiplet),
7.25 (1H, triplet, J=8Hz),
7.46-7.52 (5H, multiplet),
7.60 (1H, triplet, J=7.4Hz),
8.36 (1H, doublet, J=8Hz).

EXAMPLE 35

5-Methoxy-3-benzyl-1,2,3,4,4a,4b,5,5a,6,7-decahydro-pyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one Compound (9b)

Following a procedure similar to that of Example 33, Method 1, but starting with 4.0 g (0.01 mol) of 5-methoxy-1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride (compound 7b, as obtained in Example 24) in 300 ml of diphenyl ether, the resulting dried residue from the cyclisation reaction was neutralised with a 2N aqueous solution of sodium hydrogencarbonate and extracted with diethyl ether. The resulting residue was then chromatographed on silica gel using ethyl acetate as eluent. The resulting eluate was recrystallised twice from petroleum ether (boiling point 60-80°C to afford the title compound as a white powder in a yield of 60%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

1.49-1.70 (3H, multiplet),
1.83-2.23 (4H, multiplet),
2.42 (1H, broad singlet),
2.56 (1H, multiplet),
2.67-2.91 (5H, multiplet),
3.18 (1H, multiplet),
3.38-3.51 (2H, multiplet),
3.85 (3H, singlet),
6.88 (1H, doublet of doublets, J=2.4 and 8.9Hz),
7.05 (1H, doublet, J=2.4Hz),
7.25-7.32 (5H, multiplet),
8.34 (1H, doublet, J=8.9Hz).

EXAMPLE 36

2-Benzyl-1,2,3,4,4a,5,5a,6,7,13c-decahydropyrido[4,3-c]-pyrido[1,2,3-lm]carbazol-8-one
Compound (4a)

3.5 g (0.009 mol) of 1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride (compound 3a, as obtained in Example 23) was refluxed in 150 ml of mesitylene for 7 days under a stream of argon gas. The solvent was removed by evaporation in vacuo and the resulting residue was recrystallised from isopropanol to give the product as colourless flakes in a yield of 88%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

1.49-2.02 (7H, multiplet),
2.31-2.75 (5H, multiplet),
3.00 (1H, multiplet),
3.50 (1H, doublet, J=13Hz),
3.13 (1H, doublet, J=13Hz),
4.11 (2H, multiplet),
6.90 (1H, triplet, J=7.5Hz),
7.06 (1H, doublet, J=7.5Hz),
7.19 (1H, triplet, J=8Hz),
7.25 (5H, multiplet),
8.18 (1H, doublet, J=8Hz).

EXAMPLE 37

3-Benzyl-1,2,3,4,4a,,5,5a,6,7,13c-decahydropyrido-[3,4-c]pyrido[1,2,3-lm]carbazol-8-one
Compound (8a)

The title compound was obtained in a similar manner

to that of Example 36, but starting with 1.27 g of 1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride (compound 7a, as obtained in Example 22). The yield was 79%, without recrystallisation.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

1.45-1.58 (2H, multiplet),
1.63-1.81 (2H, multiplet),
1.92-2.04 (3H, multiplet),
2.39 (1H, multiplet),
2.59 (1H, multiplet),
2.68-2.74 (2H, multiplet),
2.88 (1H, multiplet),
2.97 (1H, multiplet),
3.03 (2H, multiplet),
3.49 (1H, doublet),
3.55 (1H, doublet),
4.10 (1H, multiplet),
7.05 (1H, triplet),
7.21 (1H, triplet),
7.25-7.34 (5H, multiplet),
7.48 (1H, doublet),
8.17 (1H, doublet).

The title compound was obtained as a racemic mixture of two enantiomers. The enantiomers were separated by means of High Performance Liquid Chromatography using a chiral column under the following conditions:

Column: Chiralpak AD (trademark, purchased from Daicel chemical Industries, Ltd);
Column Size: 25 cm x 0.46 cm;
Mobile Phase: Hexane/2-propanol (isocratic run; 9 : 1) + 0.1% diethylamine;
Flow Rate: 1 ml/minute;
UV Detector: 254 nm;

The retention times of the two enantiomers were 19.0 and 23.3 minutes and, following the above conditions, about 1 mg of each pure enantiomer was obtained from 3 mg of the racemate 8a.

EXAMPLE 38

5-Methoxy-3-benzyl-1,2,3,4,4a,4b,5,5a,6,7,13c-decahydro-pyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one Compound (8b)

Following a procedure similar to that of Example 36, but starting with 0.5 g of 5-methoxy-1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridinyl-4-yl)indole (compound 7b, as obtained in Example 24, but as free base obtained by neutralisation with dilute aqueous sodium hydrogencarbonate). The resulting residue was chromatographed on silica gel using, as eluent a 4 : 1 by volume mixture of ethylacetate and hexane to give the title compound as a light yellow powder in a yield of 80%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:

1.42-1.55 (2H, multiplet),
1.61-1.70 (2H, multiplet),
1.88-2.01 (3H, multiplet),
2.38 (1H, multiplet),
2.60 (1H, multiplet),
2.82-3.01 (3H, multiplet),
3.49 (1H, doublet, J=13Hz),
3.55 (1H, doublet, J=13Hz),
3.80 (3H, singlet),
4.09 (1H, multiplet),
6.75 (1H, doublet, J=9Hz),
7.06 (1H, singlet),

7.32 (5H, multiplet),
8.10 (1H, doublet, J=9Hz).

EXAMPLE 39

1,2,3,4,4a,5,5a,6,7,13c-Decahydropyrido[2,3-c]pyrido-[1,2,3-lm]carbazol-8-one
Compound (13)

Following a procedure similar to that of Example 33 but starting from 0.5 g of 1-(4-pentenoyl)-3-(1-t-butoxycarbonyl-1,4,5,6-tetrahydropyridin-3-yl)indole (Compound 12, as obtained in Example 25), the title compound was obtained by recrystallisation of the resulting residue from a mixture of ethyl acetate, methanol and hexane as light green needles in a yield of 80%.

Nuclear Magnetic Resonance Spectrum (400 MHz, $CDCl_3$), δ ppm:

1.49-1.98 (7H, multiplet),
2.17 (1H, multiplet),
2.62-2.90 (2H, multiplet),
2.94 (1H, multiplet),
3.22 (2H, multiplet),
3.43 (1H, doublet, J=5.6Hz),
4.05 (1H, multiplet),
7.10 (1H, triplet, J=8Hz),
7.23 (1H, triplet, J=8Hz),
7.54 (1H, doublet, J=8Hz),
8.17 (1H, doublet, J=8Hz).

EXAMPLE 40

1,2,3,4,4a,4b,5,5a,6,7-Decahydropyrido[2,3-c]pyrido-[1,2,3-lm]carbazol-8-one Compound (14)

Method 1

0.10 g of 1,2,3,4,4a,5,5a,6,7,13c-decahydropyrido-[2,3-c]pyrido[1,2,3-lm]carbazol-8-one (compound 13, as obtained in Example 39) was dissolved in 25 ml of diphenyl ether containing a catalytic amount (0.08 g) of concentrated aqueous hydrogen chloride in 0.5 ml of ethanol. The reaction mixture was then heated to 90 - 95°C for 30 minutes, cooled and then diluted with diethyl ether. The resulting precipitate was collected by filtration, washed with diethyl ether and recrystallized from a mixture of ethyl acetate, methanol and hexane to give the title compound as light green needles in quantitative yield.

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 400 MHz), δ ppm:

1.54 (1H, multiplet),
1.72-1.88 (2H, multiplet),
1.93 (1H, multiplet),
2.13 (1H, multiplet),
2.21 (1H, multiplet),
2.45 (1H, multiplet),
2.84-2.96 (3H, multiplet),
3.07-3.20 (2H, multiplet),
3.30 (1H, multiplet),
3.55 (1H, multiplet),
3.89 (1H, multiplet),
7.27-7.33 (2H, multiplet),
7.69 (1H, multiplet),
8.40 (1H, multiplet).

Method 2

1.0 g of 1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride (compound 3a, as obtained in Example 23) was heated in 100 ml of diphenyl ether to 240°C for 3 hours under an argon atmosphere with no gas-flow. After this time, the reaction mixture was diluted with diethyl ether and then hydrogen chloride in diethyl ether was added. The resulting precipitate was collected by filtration, washed with diethyl ether and dried by evaporation in vacuo. 0.96 g of the product, 4-benzyl 1,2,3,4,4a,5,5a,6,7,13c-decahydropyrido[2,3-c]pyrido[1,2,3-lm]carbazol-8-one hydrochloride, was obtained as a light green powder in a yield of 96%.

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 400 MHz), δ ppm:

1.54-1.65 (1H, multiplet),
1.77-1.88 (2H, multiplet),
1.93-2.05 (2H, multiplet),
2.09-2.24 (1H, multiplet),
2.31-2.38 (1H, multiplet),
2.74-2.93 (2H, multiplet),
2.96-3.23 (3H, multiplet),
3.34-3.43 (1H, multiplet),
3.67-3.71 (1H, multiplet),
3.86 (1H, broad singlet),
4.57 (1H, doublet, J=13.4Hz),
4.66 (1H, doublet, J=13.4Hz),
7.25-7.33 (2H, multiplet),
7.47-7.52 (3H, multiplet),
7.59-7.63 (2H, multiplet),
7.67 (1H, doublet, J=7Hz),
8.40 (1H, doublet of doublets, J=7 and 1.4Hz).

0.62 g of the product obtained above was suspended,

together with 0.60 g of 5% w/w palladium-on-charcoal, in 120 ml of methanol. The resulting mixture was stirred under an atmosphere of hydrogen gas at room temperature for 50 minutes. The catalyst was then removed by filtration and the filtrate was concentrated in vacuo to give 0.42 g of a solid residue which was recrystallized from methanol as 0.14 g of colourless needles in a yield of 28%. The resulting product was determined to be identical with the compound obtained using method 1 by nuclear magnetic resonance spectroscopy and by mass spectroscopy.

DEBENZYLATION REACTIONS

EXAMPLE 41

1,2,3,4,4a,4b,5,5a,6,7-Decahydropyrido[3,4-c]pyrido-[1,2,3-lm]carbazol-8-one
Compound (10a)

General procedure

A suspension of 0.5 g (1.25 mmol) of the hydrochloride salt of 2-benzyl-1,2,3,4,4a,4b,5,5a,6,7-decahydropyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one hydrochloride (compound 9a, as obtained in Example 33) and 0.25 g of 5% w/w palladium-on-carbon in 100 ml of methanol was stirred under an argon atmosphere for 10 minutes, and subsequently under hydrogen gas at atmospheric pressure at 40°C for 2 hours. After this time, the reaction mixture was filtered to remove the catalyst and the filtrate taken to dryness by evaporation in vacuo. The residue was recrystallized from isopropanol to give the title compound as needles

in a yield of greater than 90%.

Nuclear Magnetic Resonance Spectrum (D_2O, 400 MHz), δ ppm:

1.37 (1H, multiplet),
1.51 (1H, multiplet),
2.05 (2H, multiplet),
2.21 (1H, multiplet),
2.55-2.85 (6H, multiplet),
3.07 (1H, triplet),
3.22 (1H, broad singlet),
3.35 (2H, multiplet),
7.25 (2H, multiplet),
7.55 (1H, doublet),
8.05 (1H, doublet).

EXAMPLE 42

1,2,3,4,4a,4b,5,5a,6,7-Decahydropyrido[4,3-c]pyrido-[1,2,3-lm]carbazol-8-one Compound (6a)

Following a procedure similar to that of Example 41, but starting with 0.40 g of the hydrochloride salt of 2-benzyl-1,2,3,4,4a,4b,5,5a,6,7-decahydropyrido[4,3-c]-pyrido[1,2,3-lm]carbazol-8-one hydrochloride (compound 5a, as obtained in Example 34), the title compound was obtained in a yield of 90%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

1.59-1.71 (2H, multiplet),
1.81-1.95 (2H, multiplet),
2.15 (2H, multiplet),
2.45 (1H, multiplet),
2.79-3.12 (4H, multiplet),

3.30 (1H, multiplet),
3.40 (1H, multiplet),
3.49 (1H, multiplet),
4.22 (1H, multiplet),
7.50 (2H, multiplet),
7.72 (1H, doublet),
8.36 (1H, doublet).

EXAMPLE 43

5-Methoxy-1,2,3,4,4a,4b,5,5a,6,7-decahydropyrido[3,4-c]-pyrido[1,2,3-lm]carbazol-8-one hydrochloride Compound (10b)

The title compound was obtained by following a procedure similar to that of Example 41, but starting with 0.79 g of the hydrochloride salt of 5-methoxy-3-benzyl-1,2,3,4,4a,4b,5,5a,6,7-decahydropyrido[3,4-c]-pyrido[1,2,3-lm]carbazol-8-one (compound 9b, as obtained in Example 35). The resulting residue was run through a short column of silica gel using 5% v/v ammonia in methanol as the eluent. After evaporation of solvent under reduced pressure, the residue was dissolved in a mixture of isopropanol and diethyl ether and then treated with a solution of hydrogen chloride in diethyl ether. The resulting precipitate was collected by filtration, washed with diethyl ether and dried in vacuo at 50°C for four hours to afford the title compound in a yield of 90%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 400 MHz), δ ppm:
1.60 (2H, multiplet),
2.02 (2H, multiplet),
2.18 (1H, multiplet),
2.45 (2H, multiplet),

2.70 (2H, multiplet),
2.91 (3H, multiplet),
3.21 (2H, multiplet),
3.30 (1H, multiplet),
3.80 (3H, singlet),
6.90 (1H, doublet of doublets, J=2.3 and 8.9Hz),
7.10 (1H, doublet, J=2.3Hz),
8.20 (1H, doublet, J=8.9Hz).

PREPARATION OF 1,2,3,4,6,7-HEXAHYDROPYRIDO[4,3-c OR 3,4-c]PYRIDO[1,2,3-lm]CARBAZOL-8-ONES

EXAMPLE 44

3-Benzyl-1,2,3,4,6,7-hexahydropyrido[3,4-c]pyrido-[1,2,3-lm]carbazol-8-one
Compound (20a)

1.0 g of 1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride (compound 19a, as obtained in Example 29) was heated in 100 ml of diphenyl ether to 230 - 240°C for 5 hours under a slow stream of argon. The solvent was then removed by evaporation under reduced pressure and the residue was chromatographed on a silica gel column using, as eluent, a 1 : 1 by volume mixture of ethyl acetate and toluene to give 0.51 g of the product as a crystalline solid in a yield of 56%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 400 MHz), δ ppm:
2.93 (2H, triplet, J=5.9Hz),
3.01 (2H, triplet, J=7.5Hz),
3.20 (2H, triplet, J=7.5Hz),

3.36 (2H, triplet, J=5.9Hz),
3.76 (4H, singlet),
6.93 (1H, singlet),
7.25-7.45 (6H, multiplet),
7.50 (1H, triplet, J=8Hz),
7.98 (1H, doublet, J=8Hz),
8.54 (1H, doublet, J=8Hz).

EXAMPLE 45

2-Benzyl-1,2,3,4,6,7-hexahydropyrido[4,3-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride Compound (17a)

A suspension of 0.62 g of 1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride (compound 16a, as obtained in Example 26) in 130 ml of diphenyl ether was gradually heated to 230 - 240°C under a slow stream of argon. The reaction mixture was maintained at this temperature for 1 hour, then cooled to room temperature, diluted with diethyl ether and acidified with hydrogen chloride in diethyl ether. The resulting precipitate was collected by filtration, washed with diethyl ether and dried under reduced pressure. 0.61 g of the product was obtained as a pale yellow powder in a yield of 94%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:
3.03 (2H, triplet, J=7.4Hz),
3.20-3.30 (4H, multiplet),
3.53 (2H, broad singlet),
4.56 (2H, singlet),
4.81 (2H, singlet),
7.28 (1H, singlet),
7.51 (1H, triplet, J=8Hz),

7.56-7.65 (6H, multiplet),
7.84 (1H, doublet, J=8Hz),
8.45 (1H, doublet, J=8Hz).

Following a procedure similar to that of Example 45, but using the appropriate starting materials in appropriate quantities, the following compounds of Examples 46 to 50 were obtained.

EXAMPLE 46

2-Benzyl-12-methoxy-1,2,3,4,6,7-hexahydropyrido[4,3-c]-pyrido[1,2,3-lm]carbazol-8-one hydrochloride Compound (17b)

The starting material was 960 mg of 5-methoxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)-indole hydrochloride (compound 16b, as obtained in Example 27). 920 mg of the title compound was obtained as a white powder in a 96% yield.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:
2.98 (2H, triplet, J=7.5Hz),
3.23 (2H, triplet, J=7.5Hz),
3.26 (2H, broad singlet),
3.65 (2H, broad singlet),
3.85 (3H, singlet),
4.63 (2H, singlet),
4.76 (2H, singlet),
7.10 (1H, doublet, J=1.8Hz),
7.19 (1H, doublet of doublets, J=8.9 and 1.8Hz),
7.27 (1H, singlet),
7.56-7.71 (5H, multiplet),
8.31 (1H, doublet, J=8.9Hz).

EXAMPLE 47

2-Benzyl-12-benzyloxy-1,2,3,4,6,7-hexahydropyrido-[4,3-c]pyrido[1,2,3-lm]carbazol-8-one hydrochloride Compound (17c)

The starting material was 1.06 g of 5-benzyloxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride (compound 16c, as obtained in Example 28). 0.88 g of the title compound was obtained as a white powder in a yield of 83%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.98 (2H, triplet, J=7.5Hz),
3.23 (2H, triplet, J=7.5Hz),
3.26 (2H, broad singlet),
3.66 (2H, broad singlet),
4.64 (2H, singlet),
4.76 (2H, singlet),
5.13 (2H, singlet),
7.20 (1H, doublet, J=2.3Hz),
7.26 (1H, doublet of doublets, J=8.9 and 2.3Hz),
7.28 (1H, singlet),
7.39-7.72 (10H, multiplet),
8.31 (1H, doublet, J=8.9Hz).

EXAMPLE 48

3-Benzyl-12-methoxy-1,2,3,4,6,7-hexahydropyrido[3,4-c]-pyrido[1,2,3-lm]carbazol-8-one hydrochloride Compound (20b)

The starting material was 1.21 g of 5-methoxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)-indole hydrochloride (compound 19b, as obtained in

Example 30). 1.18 g of the title compound was obtained as a white powder in a greater than 95% yield.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.99 (2H, triplet, J=7.5Hz),
3.22 (2H, triplet, J=7.5Hz),
3.58 (2H, broad singlet),
3.64 (2H, broad singlet),
3.89 (3H, singlet),
4.42 (2H, singlet),
4.47 (2H, singlet),
7.17 (1H, singlet),
7.21 (1H, doublet of doublets, J=9.0 and 2.2Hz),
7.52 (1H, doublet, J=2.2Hz),
7.54-7.60 (5H, multiplet),
8.33 (1H, doublet, J=9.0Hz).

EXAMPLE 49

3-Benzyl-12-benzyloxy-1,2,3,4,6,7-hexahydropyrido-[3,4-c]pyrido[1,2,3-lm]carbazol-8-one hydrochloride Compound (20c)

The starting material was 1.12 g of 5-benzyloxy-1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride (compound 19c, as obtained in Example 31). 1.04 g of the title compound was obtained as a white powder in a 92% yield.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.98 (2H, triplet, J=7.5Hz),
3.22 (2H, triplet, J=7.5Hz),
3.55 (2H, broad singlet),
3.63 (2H, broad singlet),

4.42 (2H, singlet),
4.47 (2H, singlet),
5.24 (2H, singlet),
7.17 (1H, singlet),
7.28 (1H, doublet of doublets, J=8.9 and 1.8Hz),
7.36-7.61 (11H, multiplet),
8.32 (1H, doublet, J=8.9Hz).

[EXAMPLE 50]

EXAMPLE 51

1,2,3,4,6,7-Hexahydropyrido[3,4-c]pyrido[1,2,3-lm]-carbazol-8-one hydrochloride
Compound (21a)

0.32 g of 3-benzyl-1,2,3,4,6,7-hexahydropyrido-[3,4-c]pyrido[1,2,3-lm]carbazol-8-one (compound 20a, as obtained in Example 44) and 0.30 g of 5% w/w palladium-on-carbon were suspended in 300 ml of methanol and 0.44 ml of 2N aqueous hydrochloric acid. The mixture was stirred under an atmosphere of hydrogen at room temperature for 2 hours. The reaction mixture was then filtered to remove the catalyst and the filtrate was taken to dryness by evaporation in vacuo. The residue was recrystallized from methanol to give 0.17 g of the title compound as colourless needles in a yield of 64%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:
2.99 (2H, triplet, J=7.5Hz),
3.22 (2H, triplet, J=7.5Hz),
3.47 (2H, triplet, J=6.1Hz),
3.61 (2H, triplet, J=6.1Hz),

4.44 (2H, singlet),
7.21 (1H, singlet),
7.49 (1H, triplet, J=8Hz),
7.57 (1H, triplet, J=8Hz),
8.03 (1H, doublet, J=8Hz),
8.34 (1H, doublet, J=8Hz).

EXAMPLE 52

1,2,3,4,6,7-Hexahydropyrido[4,3-c]pyrido[1,2,3-lm]-carbazol-8-one hydrochloride
Compound (18a)

A suspension of 0.61 g of 2-benzyl-1,2,3,4,6,7-hexahydropyrido[4,3-c]pyrido[1,2,3-lm]carbazol-8-one hydrochloride (compound 17a, as obtained in Example 45) and 0.65 g of 5% w/w palladium-on-charcoal in 120 ml of methanol was stirred under an atmosphere of hydrogen at 40°C. The reaction mixture was filtered to remove the catalyst and the filtrate was taken to dryness, by evaporation in vacuo, to yield 0.46g of product (yield of 97%). The residue was recrystallized from a 9 : 1 by volume mixture of methanol and water to give 0.16 g of the title compound as colourless needles in a yield of 33%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

3.02 (2H, triplet, J=7.4Hz),
3.21 (2H, triplet, J=6.0Hz),
3.26 (2H, triplet, J=7.4Hz),
3.53 (2H, triplet, J=6.0Hz),
4.77 (2H, singlet),
7.28 (1H, singlet),
7.52 (1H, triplet, J=8Hz),
7.61 (1H, triplet, J=8Hz),

7.93 (1H, triplet, J=8Hz),
8.40 (1H, triplet, J=8Hz).

Following a procedure similar to that of Example 52, but using the appropriate starting materials in appropriate quantities, the following compounds of Examples 53 to 57 were obtained.

EXAMPLE 53

12-Methoxy-1,2,3,4,6,7-hexahydropyrido[4,3-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride Compound (18b)

The starting material was 850 mg of 2-benzyl-12-methoxy-1,2,3,4,6,7-hexahydropyrido[4,3-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride (compound 17b, as obtained in Example 46), and 610 mg of the title compound was obtained in a 91% yield. Recrystallisation of the title compound from a 9 : 1 by volume mixture of methanol and water gave colourless needles.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:
2.98 (2H, triplet, J=7.4Hz),
3.18 (2H, triplet, J=5.9Hz),
3.23 (2H, triplet, J=7.4Hz),
3.49 (2H, triplet, J=5.9Hz),
3.90 (3H, singlet),
4.75 (2H, singlet),
7.19 (1H, doublet of doublets, J=8.9 and 2.4Hz),
7.26 (1H, singlet),
7.35 (1H, doublet, J=2.4Hz),
8.31 (1H, doublet, J=8.9Hz).

EXAMPLE 54

12-Hydroxy-1,2,3,4,6,7-hexahydropyrido[4,3-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride Compound (18c)

The starting material was 800 mg of 2-benzyl-12-benzyloxy-1,2,3,4,6,7-hexahydropyrido[4,3-c]pyrido[1,2,3-lm]carbazol-8-one hydrochloride (compound 17c, as obtained in Example 47), and 430 mg of the title compound was obtained in an 83% yield. Recrystallisation of the title compound from a 9 : 1 by volume mixture of methanol and water gave colourless needles.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.96 (2H, triplet, J=7.5Hz),
3.19 (2H, triplet, J=6.0Hz),
3.21 (2H, triplet, J=7.5Hz),
3.52 (2H, triplet, J=6.0Hz),
4.69 (2H, singlet),
7.00 (1H, doublet of doublets, J=8.8 and 1.9Hz),
7.21 (1H, doublet, J=1.9Hz),
7.24 (1H, singlet),
8.16 (1H, doublet, J=8.8Hz).

EXAMPLE 55

12-Methoxy-1,2,3,4,6,7-hexahydropyrido[3,4-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride Compound (21b)

The starting material was 932 mg of 3-benzyl-12-methoxy-1,2,3,4,6,7-hexahydropyrido[3,4-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride (compound 20b, as obtained in Example 48), and 610 mg of the title

compound was obtained in an 83% yield. Recrystallisation of the title compound from a 9 : 1 by volume mixture of methanol and water gave colourless needles.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.99 (2H, triplet, J=7.4Hz),
3.22 (2H, triplet, J=7.4Hz),
3.47 (2H, triplet, J=5.4Hz),
3.61 (2H, triplet, J=5.4Hz),
3.90 (3H, singlet),
4.40 (2H, singlet),
7.20 (1H, doublet, J=8.9Hz),
7.22 (1H, singlet),
7.52 (1H, doublet, J=1.8Hz),
8.32 (1H, doublet, J=8.9Hz).

EXAMPLE 56

12-Hydroxy-1,2,3,4,6,7-hexahydropyrido[3,4-c]pyrido-[1,2,3-lm]carbazol-8-one hydrochloride Compound (21c)

The starting material was 940 mg of 3-benzyl-12-benzyloxy-1,2,3,4,6,7-hexahydropyrido[3,4-c]pyrido[1,2,3-lm]- carbazol-8-one hydrochloride (compound 20c, as obtained in Example 49), and 340 mg of the title compound was obtained in a 56% yield. Recrystallisation of the title compound from a 9 : 1 by volume mixture of methanol and water gave colourless needles.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

2.99 (2H, triplet, J=7.5Hz),
3.23 (2H, triplet, J=7.5Hz),
3.46 (2H, triplet, J=6.2Hz),

3.57 (2H, triplet, J=6.2Hz),
4.40 (2H, singlet),
7.05 (1H, doublet of doublets, J=8.8 and 2.2Hz),
7.20 (1H, singlet),
7.44 (1H, doublet, J=2.2Hz),
8.24 (1H, doublet, J=8.8Hz).

[EXAMPLE 57]

PREPARATION OF
1-(3-AMINOPROPYL)-4,5-DIHYDROPYRIDO[1,2,3-lm]-
CARBAZOL-6-ONES

EXAMPLE 58

1-(3-Benzylaminopropyl)-4,5-dihydropyrido[1,2,3-lm]-carbazol-6-one hydrochloride
Compound (22)

A suspension of 1.21 g of 1-(4-pentynoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-3-yl)indole hydrochloride (compound 16a, as obtained in Example 26) in 180 ml of diphenyl ether was heated to 240°C under an atmosphere of argon with no gas flow. After 1 hour at this temperature, the reaction mixture was cooled to room temperature, diluted with 300 ml of diethyl ether and acidified with hydrogen chloride in diethyl ether. The resulting light green precipitate was collected by filtration, washed with diethyl ether, and dried by evaporation under reduced pressure to give 1.14g of the title compound in a yield of 94%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 90 MHz), δ ppm:

1.90-2.30 (2H, multiplet),
2.90-3.40 (6H, multiplet),
3.80 (2H, broad singlet),
4.11 (2H, triplet, J=7Hz),
6.9-7.7 (11H, multiplet),
8.14 (1H, doublet, J=9Hz),
8.43 (1H, doublet, J=9Hz),
9.51 (2H, broad singlet).

EXAMPLE 59

1-(3-Aminopropyl)-4,5-dihydropyrido[1,2,3-lm]-carbazol-6-one
Compound (23)

A suspension of 0.70 g of 1-(3-benzylaminopropyl)-4,5-dihydropyrido[1,2,3-lm]carbazol-6-one hydrochloride (compound 22, as obtained in Example 58) and 0.59 g of 5% w/w palladium-on-charcoal in 150 ml of methanol was stirred under an atmosphere of hydrogen at 40°C for 1 hour. After this time, the mixture was filtered to remove the catalyst and the filtrate was taken to dryness by evaporation in vacuo. The resulting residue was partitioned in ethyl acetate and aqueous sodium hydrogencarbonate. The organic layer was washed with brine, and concentrated in vacuo. The title compound was obtained as 0.32 g of an amorphous powder in a yield of 66%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide/D_2O, 400 MHz), δ ppm:

1.83 (2H, multiplet),
2.71 (2H, triplet, J=7.4Hz),
2.99 (2H, triplet, J=7.4Hz),

3.11 (2H, triplet, J=7.4Hz),
3.21 (2H, triplet, J=7.4Hz),
7.15 (1H, doublet, J=7.5Hz),
7.28 (1H, doublet, J=7.5Hz),
7.49 (1H, triplet, J=8Hz),
7.57 (1H, triplet, J=8Hz),
8.12 (1H, doublet, J=8Hz),
8.44 (1H, doublet, J=8Hz).

PREPARATION OF PYRIDO[4,3-c or 3,4-c]PYRIDO[1,2,3-lm]-CARBAZOL-8-ONES

EXAMPLE 60

Pyrido[4,3-c]pyrido[1,2,3-lm]carbazol-8-one Compound (24)

A suspension of 0.8 g of 2-benzyl-1,2,3,4,4a,5,5a,-6,7,13c-decahydropyrido[4,3-c]pyrido[1,2,3,lm]carbazol-8-one (compound 4a, as obtained in Example 36) and 0.8 g of 10% w/w palladium-on-carbon in 80 ml of diphenyl ether was heated to 230°C under a slow stream of argon gas for 24 hours. The reaction mixture was then cooled to room temperature, filtered and the filtrate taken to dryness *in vacuo*. The resulting residue was stirred in small quantities of methanol. The product was collected by filtration, washed with cold methanol and dried *in vacuo* to afford 0.62g of the title compound in a yield of 90%.

Nuclear Magnetic Resonance Spectrum (tetradeuterated methanol, 400 MHz), δ ppm:
6.95 (1H, doublet, J=9.6Hz),
7.64-7.74 (2H, multiplet),

8.08 (1H, doublet, J=5.8),
8.13 (1H, doublet, J=9.6),
8.31 (1H, singlet),
8.58 (1H, singlet),
8.58 (1H, multiplet),
8.62 (1H, doublet, J=5.8),
8.80 (1H, multiplet),
10.14 (1H, singlet).

EXAMPLE 61

Pyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one Compound (25)

3.03 g of 1-(4-pentenoyl)-3-(1-benzyl-1,2,5,6-tetrahydropyridin-4-yl)indole hydrochloride (compound 7a, as obtained in Example 22) was heated in 300 ml of diphenylether to 240°C under a slow stream of argon and maintained at this temperature for 2 hours. After this time, the reaction mixture was cooled to room temperature and 1.0 g of 10% w/w palladiu-on-carbon was added. The resulting mixture was heated to 240°C for 18 hours and then cooled to about 100°C and filtered to remove catalyst. The filtrate was cooled to room temperature and the resulting white precipitate was collected by filtration, washed with diethyl ether, and dried *in vacuo* at 90°C for 3 hours to afford 1.39g of the title compound in a yield of 69%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 400 MHz), δ ppm:
6.94 (1H, doublet, J=9.5Hz),
7.66-7.75 (2H, multiplet),
8.37 (1H, doublet, J=9.5Hz),
8.71-8.80 (5H, multiplet),
9.63 (1H, singlet).

EXAMPLE 62

3-Benzylpyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one bromide
Compound (26)

A suspension of 0.34 g of pyrido[3,4-c]pyrido-[1,2,3-lm]carbazol-8-one (compound 25, as obtained in Example 61) and 1.5 ml of benzyl bromide in 34 ml of acetonitrile was stirred at 70°C for 2 hours. The resulting yellow precipitate was collected by filtration, washed with small amounts of acetonitrile, and dried *in vacuo* at 60°C for 3 hours to afford 0.48 g of the title compound in a yield of 87%.

Nuclear Magnetic Resonance Spectrum (hexadeuterated dimethylsulfoxide, 400 MHz), δ ppm:

6.09 (2H, singlet),
7.02 (1H, doublet, J=9.7Hz),
7.45-7.73 (7H, multiplet),
8.44 (1H, doublet, J=9.7Hz),
8.59 (1H, doublet, J=8.0Hz),
8.75 (1H, doublet, J=7.8Hz),
8.99 (1H, singlet),
9.02 (1H, doublet, J=7.0Hz),
9.27 (1H, doublet, J=7.0Hz),
10.50 (1H, singlet).

EXAMPLE 63

6,7-Dihydropyrido[3,4-c]pyrido[1,2,3-lm]carbazol-8-one
Compound (27)

A suspension of 0.48 g of 3-benzylpyrido[3,4-c]-pyrido[1,2,3-lm]carbazol-8-one bromide (compound 26, as obtained in Example 62) and 0.41 g of 5% w/w

palladium-on-carbon in 240 ml of methanol was stirred under an atmosphere of hydrogen at 45°C for 5 hours. The catalyst was removed by filtration and the filtrate taken to dryness in vacuo. The residue was chromatographed on silica gel using, as eluent, a 95 : 5 by volume mixture of ethylacetate and methanol to give 0.12 g of the product as a white powder in a yield of 41%.

Nuclear Magnetic Resonance Spectrum ($CDCl_3$, 90 MHz), δ ppm:

3.03-3.19 (2H, multiplet),
3.37-3.54 (2H, multiplet),
7.43-7.66 (2H, multiplet),
7.78 (1H, singlet),
8.28-8.39 (2H, multiplet),
8.58-8.71 (2H, multiplet),
9.29 (1H, singlet).

PREPARATION OF QUATERNARY STRYCHNINE DERIVATIVES

General Method A

In order to obtain the desired quaternary derivatives of strychnine, this method involves the addition of a solution of 1.5 equivalents of the halide of the desired quaternary grouping in chloroform to a solution of 1 equivalent of strychnine, also in chloroform. The reaction mixture is then stirred at room temperature or at an elevated temperature until precipitation occurs. The product is then filtered off, washed with chloroform and evaporated to dryness in vacuo. The products are obtained in substantially quantitative yields.

Following the above procedure, the following quaternary strychnine derivatives were prepared, and these were characterised by their High Performance Liquid Chromatography (HPLC) retention times (r.t.), using the the following HPLC conditions:

HPLC column: 4 mm (ID) x 250 mm Lichrosorb (trademark) RP-select B (5 μm).

Solvent system - a gradient run from 25% v/v methanol/water to 75% methanol/water over 17.5 minutes and then to 100% ethanol over the next 7.5 minutes;

Flow rate; 1 ml/min;

UV Detector; 254 nm.

EXAMPLE No.	COMPOUND NAME	RETENTION TIME (mins)
64	Strychnine	9.47
65	N-Methyl strychnine iodide	9.41
66	N-Ethyl strychnine bromide	10.13
67	N-Propyl strychnine bromide	11.53
68	N-Allyl strychnine iodide	10.29
69	N-Propargyl strychnine bromide	9.49
70	N-Cyanomethyl strychnine bromide	8.85
71	N-Benzyl strychnine bromide	13.83
72	N-Phenylethyl strychnine bromide	16.06
73	N-3-Nitrobenzyl strychnine bromide	13.83
74	N-4-Nitrobenzyl strychnine bromide	13.87
75	N-3-Propylphthalimido strychnine bromide	13.85
76	N-4-Azidophenacyl strychnine bromide	16.09
77	N-Phenacyl strychnine bromide	14.42

General Method B

This second general method for the preparation of quaternary derivatives of strychnine involves the

addition of 1.5 equivalents of solid sodium iodide and a solution of 1.5 equivalents of the halide of the desired quaternary grouping in methanol to a solution of 1 equivalent of strychnine, also in methanol. The reaction mixture is then stirred at room temperature until precipitation occurs. The product is collected by filtration, washed with small amounts of methanol and dried by evaporation in vacuo. The products are obtained in a greater than 80% yield. The products were characterised by HPLC under the same conditions as for Examples 64 to 77 above.

EXAMPLE No.	COMPOUND NAME	RETENTION TIME (mins)
78	N-Cyclohexylmethylstrychnine iodide	14.69
79	N-2-naphthylmethylstrychnine iodide	17.85
80	N-4-biphenylmethylstrychnine iodide	19.38

EXAMPLE 81

N-2-Carboxyethyl strychnine

A solution of 0.32 g (0.0044 mol) of beta-propiolactone in 30 ml of dimethylformamide was added dropwise with stirring to a solution of 1.0 g (0.003 mol) of strychnine in 30 ml of dimethylformamide. The reaction mixture was stirred at room temperature for one hour and the product was filtered off, washed with dimethylformamide and dried at 60°C at a pressure of 0.1 mmHg. The resulting compound had a retention time of 8.17 minutes, when characterised by HPLC under the same conditions as for Examples 64 to 77 above.

EXAMPLE 82

N-3-Sulphonylpropyl strychnine

A solution of 0.55 g (0.0045 mol) of 1,3-propane sulfone in 30 ml of dimethylformamide was added dropwise with stirring to a solution of 1 g (0.003 mol) of strychnine in 30 ml of dimethylformamide . The reaction mixture was stirred at room temperature for one hour and the product was filtered off, washed with dimethylformamide and dried at 60°C at a pressure of 0.1mmHg. The resulting compound had a retention time of 6.92 minutes, when characterised by HPLC under the same conditions as for Examples 64 to 77 above.

PREPARATION OF QUATERNARY BRUCINE DERIVATIVES:

General Method A

A similar method is followed as that described for the preparation of quaternary strychnine derivatives under Method A, except that a solution of brucine in chloroform is used. The products are usually obtained in greater than 80% yields. Following this procedure, the compounds of the following Examples were obtained and characterised by HPLC under the same conditions as for Examples 64 to 77 above.

EXAMPLE No.	COMPOUND NAME	RETENTION TIME (mins)
83	Brucine	10.13
84	N-Methyl brucine iodide	10.95
85	N-Chloromethyl brucine chloride	10.05
86	N-Allyl brucine iodide	10.41

87	N-Benzyl brucine iodide	13.17
88	N-4-Azidophenacyl brucine bromide	15.98
89	N-Phenacyl brucine bromide	14.07

General Method B

This second general method for the preparation of quaternary brucine derivatives requires the addition of 0.33 g (0.0013 mol) of silver triflate and a solution of the respective halide in 5 ml of acetone to a solution of 0.5 g (0.0013 mol) of brucine in 10 ml of acetone. The reaction solution is stirred at 50°C for 4 hours. The product is then collected by filtration, washed with small amounts of methanol and dried by evaporation *in vacuo*. The products are obtained in about 20% yield.

EXAMPLE 90

N-2-Nitrobenzyl brucine triflate

Following the above General Method B, the title compound was obtained, having a retention time of 14.93 minutes, when characterised by HPLC under the same conditions as for Examples 64 to 77 above.

M&C FOLIO: 230P70859/FP-9414 WANGDOC: 1008D

The compounds of the present invention may be assayed for allosteric activity at muscarinic receptors by a number of methods. Suitable such methods are described below, but it will be understood that other methods may be employed by those skilled in the art, and that none of the methods below, individually, is absolutely essential to the establishment of activity of the compounds of the invention.

While we generally prefer to perform all of the assays described below for each individual compound, it will be understood that this may be unnecessary where, for example, the primary assay yields a result which is not compatible with the effect required. Carrying out the remainder of the assays is generally desirable to confirm the results of the primary assay, and to provide a more detailed interpretation of the results obtained in the primary assay.

We prefer to carry out the primary assay first, as it is generally the most accurate, but this is by no means essential, and it may be preferable, depending on the circumstances, to carry out the functional (GTP assay) first, for example.

It will be understood that the present invention also envisages any and all of the accompanying assays, as described below, as well as any compounds, and the use of any compounds, which exhibit an allosteric effect by any one or more of such assays.

In the following assays, it is necessary, or at least desirable, to use a cell line which expresses only one type of human muscarinic receptor, such as m1, and which does not exhibit a high level of

acetylcholinesterase activity.

A suitable cell line is CHO (Chinese Hamster Ovary), which are readily engineered to express only one receptor sub-type.

Preparation of CHO cell membranes

To obtain the large amount of cell membranes required, plates of 530 cm^2 culture area were used. CHO cells which express human m1, m2, m3 and m4 receptors were grown separately in MEM alpha medium containing 10% newborn calf serum and antibiotics. When cells reached confluence, they were washed twice with 10 ml of 20 mM HEPES containing 10 mM EDTA (pH 7.4), scraped into the same buffer and homogenized using a Polytron (trademark) homogenizer (setting 5-6 for 5 sec x 2). Membrane pellets were obtained by centrifugation (40000xg, 10 min, 4°C) and resuspended in 20 mM HEPES - 0.1 mM EDTA (pH 7.4). Centrifugation and resuspension were repeated twice to wash the cell membranes. After measurement of membrane protein, the membranes (1 or 2 mg protein/ml) were stored at -70°C.

Protein Assay

Membrane protein was measured with Bio-Rad protein reagent using BSA (bovine serum alumin) as standard.

Assessment of cholinesterase activity

Cholinesterase activity in native CHO cell membranes was measured by the method of Ellman et al. [Biochem. Pharmac., 7, 88 (1961)] with slight modifications. Briefly, membranes were added to the solution which contained acetylthiocholine (0.6 mM) and DTNB (0.5 mM) in 20 mM HEPES - 1 mM $MgCl_2$ (pH 7.4), and absorbance

at 412 nm was measured up to 10 min. The mean rate of change of absorbance, the index of cholinesterase activity, was calculated.

If CHO cell membranes had cholinesterase activity, it would be difficult to detect ^{3}H-ACh binding and it would be necessary to block the cholinesterase activity with an inhibitor (e.g. neostigmine). Therefore, cholinesterase activity in native CHO cell membranes was assessed before examining acetylcholine (ACh) binding.

The rate of change of absorbance at 412 nm was the same with or without native CHO cell membrane (basal 0.0022/0.0010, 50 μg protein/ml 0.0030/0.0004; 100 μg/ml 0.0022/0.0010). Thus, there was no detectable cholinesterase activity in native CHO cell membranes. When rat heart membranes were used, cholinesterase activity was readily measured (> 10 times basal level at 20 μg protein/ml). Furthermore, the inhibitory effect of ACh on ^{3}H-OxoM binding to m2 membranes was not enhanced by 2.5 μM neostigmine.

Functional Assay (GTP)

In general, the final test of positive allosterism with ACh is the functional assay. We use measures of G-protein activation in membrane preparations - the GTPase assay using ^{32}P-GTP, and the ^{35}S-GTPγS binding assay. Both assays provide a true measure of of function, but the latter assay is generally easier to use and more accurate. The rate of ^{35}S-GTPγS binding reflects the ability of the agonist to reduce GDP affinity for the G-protein, which is how agonists at G-protein- coupled receptors work. A positive effect in at least one of these assays confirms positive allosterism with ACh.

The assay buffer used was 20 mM HEPES + 100 mM NaCl + 5 (GTPase) or 10 (GTPγS binding) mM $MgCl_2$. Buffers were pH7.4.

For the GTPase assay, 5 μg protein was incubated in buffer + 1 mM ATP + [γ^{32}P]GTP (10-100 nM or as indicated) + drugs in a volume of 100 μl at 30°C for 15 minutes. The reaction was stopped by addition of 750 μl of a slurry of 5% charcoal in 20 mM H_2PO_3 + 1 mg/ml BSA. After centrifugation (20,000xg, 5 mins) an aliquot of the supernatant, containing released labelled phosphate, was counted for radioactivity.

For the GTPγS assay, 15 - 50 μg membrane protein was incubated in buffer + GDP + (0.1 μM for m1 and m3 receptors, 1 MM for m2 and m4 receptors, or as indicated) + [^{35}S]GTPγS (50-100 pM) + drugs in a volume of 1 ml at 30°C for 30 minutes. Bound label was collected by rapid filtration using a Brandel (trademark) cell harvester and counted for activity.

Figure 0 shows an example of a positive allosteric agent (chloromethyl brucine) stimulating the potency of ACh in the ^{35}S-GTPγS binding assay using membranes containing m3 receptors.

Equilibrium Radioligand Binding Assays

Introduction

The binding of ACh can be measured in two ways: directly, using ^{3}H-ACh, and indirectly using the inhibition by cold ACh of ^{3}H-NMS binding. Such studies give different results, but they can be reconciled, as we have established that there are at least two components of agonist binding - a high and a

low affinity component. ^{3}H-ACh will bind only to the high affinity component, and the size of this component depends on the guanine nucleotides and divalent cations in the assay. An increase in the apparent affinity of ACh for either or both components may produce the necessary functional increase in effectiveness.

Primary Screen

There are two parts to the screen, and either may be used, or both together, although the second part is generally restricted in usefulness to m2 and m4 receptors. The first part of the screen measures the effect of unknown agent on ^{3}H-NMS binding in the absence and presence of cold ACh (GTP is present to prevent high affinity agonist binding).

The second part of the primary screen involves binding of ^{3}H-ACh to m2 and m4 receptors (^{3}H-ACh does not bind well to m1 or m3 receptors).

Procedure

Membranes are incubated in 1.12 ml (^{3}H-NMS) or 0.25 ml (^{3}H-ACh) of buffer containing 20mM HEPES + 100 mM NaCl + 10 mM MgCl2 (+0.2 mM GTP in ^{3}H-NMS assays), pH 7.4, at 30°C for two hours. The concentration of membranes is chosen to allow detectable binding of ^{3}H-ACh and ^{3}H-NMS while minimising depletion of free ^{3}H-NMS (up to about 15% depletion is acceptable) - about 100 µg protein/tube for m2, about 400 µg protein/tube for m4 membranes are used in the ^{3}H-ACh assay, 5 µg/ml of m1 membranes and 8 µg/ml of m3, 30 µg/ml of m2 membranes and 50 µg/ml of m4 membranes are used in the ^{3}H-NMS assays. The bound radioligand is collected by filtration through Whatman GF/B glass-fibre filters soaked in 0.1% polyethylenimine

using a 30-place Brandel cell harvester, and the radioactivity measured with liquid scintillation counting. Nonspecific binding is measured in the presence of 1 μM QNB.

Design and analysis

The ^{3}H-ACh assay uses a concentration of about 1 nM (m2) or 4 nM (m4) and measures total binding, nonspecific binding and binding in the presence of three concentrations of each of four agents, all in duplicate (quadruplicate for total binding).

The ^{3}H-NMS assay contains 0.1 mM GTP and uses ^{3}H-NMS concentrations of about 4 and 0.5 nM (0.4 nM with m2). Fixed ACh concentrations are 2 μM (m2), 5 μM (m4) or 20 μM (m1 and m3). Total and nonspecific binding are measured with 4 nM ^{3}H-NMS to allow an estimate of B_{max}. Using 0.2 nM ^{3}H-NMS, binding in the absence and presence of ACh is measured alone and in the presence of three concentrations of each of four agents, and nonspecific binding is measured with QNB alone. Each point is measured in duplicate (quadruplicate for 0.2 ^{3}H-NMS alone).

The data are analyzed as described below, and graphs produced, using the Minitab program. Where possible, IC_{50} values are estimated visually from the graphs.

Calculations

A plot is required, showing the relative affinity of the ligand (^{3}H-NMS or ACh) in the presence of various concentrations of the agent.

Results from the main part of the primary screen are analysed by converting each data point into an apparent

affinity. We use 5 control points - total and non-specific binding with a high and a low concentration of ^{3}H-NMS and the low ^{3}H-NMS concentration with cold ACh (at around its IC_{50}). 2-Point Scatchard analysis of specific binding with the high and low ^{3}H-NMS concentrations provides an estimate of B_{max} (total receptor concentration) and ^{3}H-NMS Kd. Binding is measured with 3 concentrations of test agent, both in the absence and presence of ACh. Given the Bmax already estimated, the amount of binding in the presence of agent alone is transformed to an estimate of ^{3}H-NMS Kd in the presence of agent. Binding in the presence of agent + ACh is related to binding in the presence of agent alone to estimate the IC_{50} of ACh (approximating that ACh binds with a Hill slope of 1), and this is further transformed to an estimate of ACh Kd using the known concentration of ^{3}H-NMS, the estimated Kd of ^{3}H-NMS in the presence of agent, and the Cheng-Prussof correction:

$$(ACh\_Kd = IC_{50} / (([^3H\text{-}NMS] / {}^3H\text{-}NMS\_Kd) + 1)$$

Finally, the affinity (1/Kd) estimates of both ^{3}H-NMS and ACh are expressed as a fraction of the corresponding control Kd in the absence of agent.

It can be shown that the concentration effect curve of an agent for this "relative affinity" measure corresponds to the agent's occupancy curve for the receptor and, if the agent is inhibitory, its IC_{50} corresponds to its Ki (1/affinity).

In the second part of the primary screen, specific binding of ^{3}H-ACh is expressed (i.e. after subtraction of non-specific binding and in the absence of GTP) as a fraction of control specific binding. Generally effects on ^{3}H-ACh binding are similar to those on cold ACh affinity, and are interpreted in the same way, but sometimes an increase in cold (non-labelled) ACh

affinity is not paralleled by an increase in ^{3}H-ACh binding. This pattern indicates that the increase in cold ACh affinity may be an artefact. The agents which are genuinely positive with ACh at m2 receptors or m4 receptors increase ^{3}H-ACh binding. A very large increase in ^{3}H-ACh binding may be an artefact.

Interpretation of Primary Screen Results

Examples of the types of result that can be expected are given below:

a) The agent reduces the apparent affinity of ^{3}H-NMS and ACh equally, with almost complete inhibition over a concentration range of 2 log units. This pattern either indicates that the agent acts competitively, or with high negative allosterism with both ^{3}H-NMS and ACh. The concentration of agent which reduces ligand affinity by 50% is the Kd of the agent for the free receptor.

b) The agent reduces the apparent affinity of ^{3}H-NMS and ACh equally, but to a limited extent. This suggests that the agent might be acting allosterically with low negative allosterism. The concentration of agent which causes 50% of its own maximal effect approximates to the Kd of the agent for the free receptor.

c) The agent inhibits the affinity of both ligands almost completely, but with different apparent potency with each ligand. This indicates that the agent is acting allosterically but with different degrees of negative allosterism with the two ligands. The 50% effective concentration of agent in the more potent curve approximates to the Kd of the agent for the free receptor.

d) The agent increases the apparent affinity of

^{3}H-NMS. This indicates an allosteric action, and this interpretation can be confirmed using the 'off-rate' assay (below).

e) The agent causes a small increase in ACh affinity with little effect on ^{3}H-NMS. This may indicate allosterism, but other assays are needed to confirm the positive effect with ACh.

f) The agent causes an increase in ACh affinity and a large decrease in ^{3}H-NMS affinity. This may indicate allosterism. If the agent has no effect in the off-rate assay then it is probably allosteric and positive with ACh.

g) The agent has no effects. Either it is inactive, or allosteric but neutral. This is readily checked in an 'off-rate' assay, and if the agent is allosteric then its affinity for the free receptor is the same as its affinity for the ^{3}H-NMS-occupied receptor.

Secondary Binding Assay

This assay provides an estimate of the affinity of an allosteric agent at the unoccupied receptor and its allosteric effect on the radioligand (usually ^{3}H-NMS) and a second unlabelled muscarinic drug (usually ACh). The assay consists of four inhibition curves with the muscarinic drug, alone and in the presence of three concentrations of the allosteric agent, against a low concentration of radioligand. Binding of a high concentration of radioligand is also measured and used to estimate the B_{max} (total receptor number) in the membranes. The complete data set is fitted simultaneously with a non-linear curve-fitting programme (SigmaPlot) to the allosteric model. When the competitive agent is ACh or another agonist, the assay

contains 2 x 10^{-4}M GTP or GppNHp.

This assay allows effects seen in the primary screen to be studied more rigorously and accurately, and provides quantitative estimates of the agent's affinity for the free receptor and maximal allosterism with both ^{3}H-NMS and ACh (from which its affinity for the ^{3}H-NMS- and ACh-occupied receptors can be calculated). Again, the results must be compatible with primary and off-rate assays. A complication is that effective concentrations of agents which are allosterically neutral or positive with ^{3}H-NMS may produce kinetic artefacts, so data obtained with the highest concentration of agent may be misleading, and a data set which is not well fitted to the model should be reanalysed without the top concentration data.

Non-equilibrium ^{3}H-NMS Binding Assay

"Off-Rate" Assay

The 'off-rate' assay measures the affinity of the test agent for the ^{3}H-NMS-occupied receptor.

The method for the 'off-rate' assay generally involves making the test drugs up to their final concentration (usually four concentrations of three drugs) in 1 μM QNB and 100 μl is distributed to tubes, except for some tubes which have no addition and some which receive QNB alone. A high concentration of ^{3}H-NMS (about 5 nM total final concentration, though a large fraction of this is bound to receptors, resulting in very low non-specific binding) is added to a 10-fold greater volume of concentrated membrane preparation and incubated for about 15 minutes. 10 μl of this preparation is added to the tubes (separate tubes are prepared with 10 μl membrane + 10 μM QNB, a potent

antagonist, followed by 1 µl ^{3}H-NMS, to measure non-specific binding). About two half-lives later (m1 20 min, m2 6 min, m3 and m4 25 min) the assay is terminated by filtration. The control dissociation constant (K_{off}) is estimated by

$$[\ln(B_0/B_t)]/\text{time}$$

where B_0 is control binding (without dissociation) and B_t is the binding remaining after addition of QNB alone. The dissociation rates in the presence of each concentration of allosteric agent are estimated in the same way, and the % inhibition of dissociation is calculated. The % inhibition values are fitted with non-linear regression to a logistic model with a slope of 1, if appropriate, to yield a pEC_{50} (pKd at occupied site) and E_{max} (maximal inhibition of dissociation rate). The E_{max} usually, but not always, approaches 100%.

The results of this assay should normally be compatible with those of the primary screen, but may alter the interpretation of the primary screen results. Typical examples are:

a) The agent inhibits ^{3}H-NMS binding in the primary screen with an apparent pKd of 4.5, the highest concentration testable is 10^{-4}M, and the agent has no effect on ^{3}H-NMS dissociation. These results are compatible with both a competitive mode of action and negative allosteric mode of action - if the agent is allosteric then its potency at the ^{3}H-NMS occupied site may be too weak for even the highest testable concentration to have any effect.

b) The agent inhibits ^{3}H-NMS binding with an apparent Kd of 10^{-5}M, and has a pKd of 5.5 in the off-rate assay. The agent is probably allosteric but neutral with ^{3}H-NMS, and the inhibition seen in the primary

screen is a kinetic artefact.

c) The agent increases ACh affinity in the primary screen with little effect on ^{3}H-NMS, but it shows activity in the off-rate assay at slightly lower concentrations than those increasing ACh affinity. The agent probably is an allosteric enhancer of ACh affinity.

Value and validation of 'off-rate' assay

This assay has three main uses:
1) It provides an estimate of the affinity of an allosteric agent for the ^{3}H-NMS occupied site, which may be useful in structure/activity considerations.
2) It provides an estimate of the affinity of an allosteric agent for the unoccupied site if the agent is 'neutral' or has only a small allosteric effect.
3) It indicates the concentration range in which kinetic artefacts may occur i.e. inhibition of ^{3}H-NMS binding due to a failure of the reaction to reach equilibrium, rather than through allosteric or competitive inhibition (the time to reach equilibrium is directly related to the dissociation rate).

It is necessary to show that:
1) ^{3}H-NMS dissociation alone and in the presence of an allosteric agent follows a mono-exponential time course, so that the rate constants can be measured from a single point,
2) dissociation rate constants in the presence of various concentrations of allosteric agent fit a single-site model (logistic function with a slope of 1), consistent with binding of the agent according to the Law of Mass Action,
3) the parameters obtained with the assay are independent of the dissociation time at which binding is measured, and

4) the parameters are consistent with known results.

Dissociation of ^{3}H-NMS was measured at m1-m4 sites at 1, 2 and 4 half lives, alone and in the presence of three concentrations of strychnine, an agent which we have established is an allosteric agent with small effects on equilibrium ^{3}H-NMS binding.

Figure 1 shows the data plotted against time and well fitted to a monoexponential function, thus fulfilling the first criterion.
Figure 2 shows the rate constants, measured from the fits shown in Figure 1, plotted against strychnine concentration and well fitted to a single-site model, thus fulfilling the second criterion.
Figure 3 shows, for m4 data, how the dissociation data with different strychnine concentrations can be treated as dose-response data at different times. Scaling the data by plotting the inhibition of the counts (dpm) dissociated in the absence of agent causes the effect of strychnine to appear to be weaker when measured at 40 minutes than at earlier times but, when the data are converted to off-rates, as described above, then the potency of strychnine is independent of time.

This is illustrated in Figure 4, where pIC_{50} values, measured either from untransformed dpm data or from the rate-constant transformation, are plotted against time for m1-m4. pIC_{50}s measured from dpm data decrease with time, whereas pIC_{50}s measured from transformed data are essentially independent of time. This result fulfils the third criterion.

Strychnine has positive or small negative allosteric effects, but in a secondary screen with m2 receptors a pK of 4.7 was estimated for strychnine at the free receptor, and 2-fold positive allosterism with ^{3}H-NMS,

which is entirely consistent with the pK of 5.1 at the occupied receptor estimated in the above experiment.

Thus, this experiment provides firm validation for the single-point off-rate screening assay.

The results of the above tests are presented below for various compounds of the present invention as Tables 1, 1a, 2, 2a, 3 and 3a. Compound numbers correspond to those in the Tables of formulae above. Tables "X" relate to the primary screen, the off-rate screen and, where appropriate, the functional (GTP) assay. Tables "Xa" relate to the secondary screen.

In the following Tables, the results are interpreted as follows:

Primary screen

a) ^{3}H-NMS binding

++	large increase
+	clear increase
(+)	small increase, may be noise
0	inactive or neutral
(-)	small inhibition, drug is weak, non-specific effect, or noise
-	clear inhibition, may be allosteric
--	strong inhibition up to 100%, competition or large negative cooperativity
(--)	strong inhibition, could be kinetic artefact

b) Unlabelled ACh

+++	huge increase in apparent affinity, probably artefact
++	large increase in apparent affinity
+	clear increase
(+)	small increase, could be noise
0	inactive or neutral
(-)	small inhibition, agent is weak, or noise
-	inhibition, could be allosteric
--	large inhibition, competitive or strong negative cooperativity

c) 3H-ACh

+++	Huge effect, probably artefact
++	Large positive effect
+	Clear positive effect
(+)	small positive effect, could be noise or positive allosterism
0	Inactive or neutral
(-)	Small negative effect, could be noise, lack of potency or small negative allosterism
-	Clear inhibition
--	Large negative effect, up to 100% inhibition.

'Off-rate' screen

0	no effect
-	small inhibition at highest concentration, maybe non-specific
(pK)	partial inhibition, pK may be inaccurate
pK	strong inhibition, reliable pK
(pk)*	maximal inhibition <80%

Secondary screen

0 no effect.

If there is a quantifiable effect, three values are given:

pK	the -log Kd of the agent for the free receptor.
αNMS	the maximal allosterism (fold change in affinity) with 3H-NMS.
αACh	the maximal allosterism with ACh

Small values of α, e.g. <0.01, which are not distinguishable from 0 are given as 0.

Any value with poor statistical precision, either because the effect is small or the fit is poor, is enclosed in brackets ().

Functional GTPase or ^{35}S-GTPγS binding assay

Qualitative result:

++	large increase in ACh potency
+	clear increase in ACh potency
0	no effect
-	clear decrease in ACh potency
--	large decrease in ACh potency
( )	imprecise/small effect

Quantitative result:

pK	- log apparent Kd for free receptor
α	maximal allosterism with ACh: small values, e.g. <0.01, which are not distinguishable from 0 are given as 0.
()	imprecise/small effect

SMB

-	SMB (sodium metabisulphite) abolished a positive effect on ACh
(-)	SMB reduced a positive effect on ACh
0	SMB had no effect
(+)	SMB may have revealed a positive effect on ACh

TABLE NO. 1

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
001	m1	0	--		4.9	--	
	m2	++	--	--	5.2		
	m3	0	--		4.3		
	m4	+	-	-	5.1		
002	m1	-	-		4.2	(-)	
	m2	-		--	4.6		
	m3	-	-		3.9		
	m4	-	-	-	4.3		
003	m1	-	-		(3.5)	(-)	
	m2	-		-	(3.7)		
	m3	-	-		(3.3)		
	m4	(-)	-	-	(3.6)		
004	m1	-	-			(-)	
	m2	-		-			
	m3	(0)	(0)				
	m4	0	0				
005	m1	-	-			(-)	
	m2	-		--			
	m3	-	-				
	m4	-	-	-			
006	m1	--	-			(-)	
	m2	-		--			
	m3	(-	-)				
	m4	-	-	-			
007	m1	--	-		4.0	-	
	m2	--		--	4.7	-	
	m3	-	-		3.8	-	
	m4	-	-	--	4.2		
008	m1	-	-			(-)	
	m2	(-)		-			
	m3	(-)	(-)				
	m4	-	--	--			
009	m1	--	--		(4.0)	(-)	
	m2	-		--	4.9		
	m3	n o d a t a			(3.5)		
	m4	-	-	-	4.3		
010	m1	-	-			(-)	
	m2	-		-			
	m3	(-)	(0)				
	m4	(-)	(-)	-			

TABLE NO. 1 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
011	m1	-	-		4.4	-	
	m2	++		--	6.5	-	
	m3	(-)	(-)		3.5	-	
	m4	0	-	-	4.7		
012	m1	n o d a t a				-	
	m2	-		--			
	m3	n o d a t a					
	m4	n o d a t a					
013	m1	0	-		4.4	-	
	m2	+	-	-	5.0		
	m3	+	-		4.1		
	m4	+	-	-	4.7		
014	m1	-	--		4.6	-	
	m2	+		-	5.2		
	m3	(-)	-		4.4		
	m4	(+)	--	--	5.0		
015	m1	--	--		4.8	-	
	m2	+		-	5.4		
	m3	-	-		4.4		
	m4	-	-	--	4.9		
016	m1	--	--			-	
	m2	-		--			
	m3	0	-				
	m4	-	-	-			
017	m1	-	-		4.7	-	
	m2	0		-	5.3		
	m3	-	-		4.5		
	m4	0	-	--	5.0		
018	m1	0	-		4.5	-	
	m2	++		-	5.0		
	m3	+	-		4.1		
	m4	++	-	-	4.7		
019	m1	0	-		4.7	-	
	m2	++		--	5.4	-	
	m3	+	-		4.2	-	
	m4	(0)	(-)	(-)	4.7		
020	m1	0	-		5.3	-	
	m2	++	-	-	6.3		
	m3	0	-		5.4		
	m4	(0)	-	-	5.7		

TABLE NO 1 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
021	m1	-	-		4.8		
	m2	+		-	5.7		
	m3	0	-		4.5		
	m4	0	-	-	5.5		
022	m1	+	-		5.0	?low neg coop with ACh	
	m2	++	-	-	5.9		
	m3	+	-		4.8		
	m4	0	-	-	5.5		
023	m1	+	-		5.1	-	
	m2	++	-	-	5.5		
	m3	-	-		4.5		
	m4	0	-	-	5.3		
024	m1	-	-			(-)	
	m2	+		-			
	m3	+	-				
	m4	0	-	-			
025	m1	0	-		5.2		
	m2	+		-	5.8		
	m3	(0)	-		5.7		
	m4	+	(-)	(-)	6.3		
026	m1	0	-		-	(0)	
	m2	+		-	-		
	m3	+	0		-		
	m4	++	(0)	(0)	-		
027	m1				-		
	m2				-		
	m3				-		
	m4				-		
028	m1	+	(=)		4.0		
	m2	+	0	(-)	4.3		
	m3	++	(-)		4.0		
	m4	+	0	0	4.2		
029	m1	0	-		4.6	-	
	m2	++	-	-	5.3	-	
	m3	0	-		4.3	-	
	m4	+	-	-	4.8	-	
033	m1	-	+		4.6	Potentiation found in 7expts	
	m2	+	-	-	4.6		
	m3	(-)	(0)		3.5		
	m4	0	0	0	4.7		

TABLE NO. 1 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
034	m1	-	-		4.0		
	m2	+	-	-	4.6		
	m3	(-)	(+)		3.6		
	m4	0	-	-	4.5		
035	m1	-	0		4.2		
	m2	+	-	-	4.7	-	
	m3	0	++		3.6	+/+	
	m4	+	(+)	(+) (-)	4.5	+	
036	m1	(0)	(+)			--/--	
	m2	--	++	--			
	m3	0	++				
	m4	0	0	(-)		--	
037	m1	(+)	(0)		3.4	/0	
	m2	+	-	-	3.5	(-)	
	m3	+	+		3.4	/+	
	m4	+	+	+	3.7	+/+	
038	m1	(-)	(-)			/0	
	m2	+	0	(-)			
	m3	(0)	+				
	m4	0	-	-			
039	m1	-	(+++)			--	-
	m2	-	(+++)	-			-
	m3	-	(+++)			/(--)	-
	m4	--	+	++			-
040	m1	-	-				
	m2	+	-	-			
	m3	-	+				
	m4	-	-	-			
055	m1	+	-				
	m2	++	-	-			
	m3	0	-				
	m4	++	-	--			
056	m1	(0)	-				
	m2	0	-	-			
	m3	-	-				
	m4	(-)	-	-			
057	m1	-	-				
	m2	0	-	-			
	m3	-	-				
	m3	0	-	-			

TABLE NO. 1 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
058	m1	(0)	-				
	m2	(0)	-	-			
	m3	(-)	(-)				
	m4	(0)	-	-			
059	m1	-	-				
	m2	-	-	-			
	m3	-	-				
	m4	-	-	--			
060	m1	-	--				
	m2	-	--	-			
	m3	-	-				
	m4	0	-	--			
061							
062	NO DATA						
063							
064	m1	-	-				
	m2	+	0	-			
	m3	0	0				
	m4	+	+	(-)			
065	m1	-	0				
	m2	+	-	-			
	m3	0	0				
	m4	+	(+)	-		/-	
076	NO DATA						
188	m1	+	-		5.2		
	m2	+	--	--	5.8		
	m3	-	--		5.2		
	m4	-	--	--	5.5		
214	m1	0	(-)		3.8		
	m2	+	-	-	4.2		
	m3	(+)	(-)		3.8		
	m4	+	-	-	4.2		

TABLE NO. 1a

Compound no.	Receptor	SECONDARY ASSAY pK	%NMS	%ACh
033	m1	(3.7)	0.5	1.9
	m2			
	m3			
	m4			
035	m1	(3.2)	(0.5)	0.7
	m2	4.6	1.3	0.2
	m3	3.4	0.3	4.9
	m4			
038	m1			
	m2			
	m3	2.9	0.14	3.8
	m4			
				+SMB
039	m1	5.1	0.6	1.1
	m2			
	m3			
	m4			
040	m1			
	m2			
	m3	3.8	0.04	2.4
	m4			
064	m1			
	m2			
	m3			
	m4			+ve
065	m1			
	m2			
	m3			
	m4	5.0	1.5±0.2	1.9±0.5

TABLE NO. 2

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
099	m1	--	--				
	m2	-	--	--			
	m3	--	--				
	m4	(-)	--	--			
100	m1	++	0		4.8		
	m2	++	0	0	4.4		
	m3	+	0		4.1		
	m4	++	0	0	4.6		
101	m1	--	--		3.9		
	m2	--	--	--	3.6		
	m3	--	--		3.7		
	m4	--	--	--	4.0		
102	m1	--	--				
	m2	(-)	--	--			
	m3	--	--				
	m4	(-)	--	--			
103	m1	0	--				
	m2	+	-	-			
	m3	(0)	0				
	m4	++	-	-			
104	m1	0	-				
	m2	++	0	0			
	m3	++	(0)				
	m4	(+)	(-)	--			
105	m1	(-)	--				
	m2	-	--	(-)			
	m3	(-)	(-)				
	m4	++	-	-			
106	m1	(+)	(-)				
	m2	++	(-)	(-)			
	m3	++	+			(-)GTPase (=)GTPγS	
	m4	++	0	(-)			
107	m1	-	--				
	m2	0	--	--			
	m3	(-)	+				
	m4	--	--	--			

TABLE NO. 2 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
113	m1	+	+				
	m2	++	-	-			
	m3	++	0				
	m4	++	0	-			
114	m1	--	--				
	m2	--	--	--			
	m3	--	--				
	m4	--	--	--			
115	m1	--	--				
	m2	--	-	--			
	m3	--	--				
	m4	--	--	--			
116	m1	-	++				small inhbtion larger wth SMB
	m2	0	++	0			
	m3	(-)	++				
	m4	(-)	++	(-)			
118	m1	--	--				
	m2	-	--	--			
	m3	--	--				
	m4	--	--	--			
119	m1	++	--				
	m2	++	-	-			
	m3	++	-				
	m4	++	-	-			
120	m1	--	-				
	m2	--	--	-			
	m3	(--)	-				
	m4	++	-	-			
121	m1	--	--		4.6		
	m2	--	--	-	4.1		
	m3	--	--		4.1		
	m4	--	--	--	4.4		
122	m1	--	--				
	m2	-	--	-			
	m3	--	--				
	m4	(-)	--	--			
123	m1	(+)	0		(4.6)*		
	m2	0	0	0	(4.2)*		
	m3	0	(-)		(3.9)		
	m4	+	0	-	(4.6)*		

TABLE NO. 2 contd

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
124	m1	++	-				
	m2	++	-	-			
	m3	++	0				
	m4	++	-	-			
125	m1	--	--				
	m2	-	-	-			
	m3	--	--				
	m4	(-)	--	--			
126	m1	0	0				
	m2	-	-	0			
	m3	(-)	(-)				
	m4	0	(-)	(-)			
129	m1	--	--				
	m2			--			
	m3	--	--				
	m4	--	--	--			
130	m1	-	-		4.7		
	m2	++	-		4.7		
	m3	-	--		4.5		
	m4	0	--		4.8		
131	m1	0	0		3.7*		
	m2	0	-	0	4.0*		
	m3	(-)	(-)		3.8*		
	m4	(0)	(-)	(-)	3.8		
132	m1	-	-		3.9		
	m2	+	-	(-)	3.8		
	m3	0	(-)		3.8		
	m4	(+)	(-)	(-)	3.9		
133	m1	0	0		3.6		
	m2	(+)	-	0	3.3		
	m3	0	0		3.8		
	m4	+	0	(-)	3.5		
134	m1	0	+		4.6*	0	
	m2	(+)	(-)	0	4.8*		
	m3	(-)	(0)		4.4*		
	m4	(+)	(0)	(0)	4.5*		
135	m1	(--)	-		5.6		
	m2	+	+	+	5.5	pk=5.1, x=7	
	m3	(+)	-		5.3	0	
	m4	+	-	-	5.7	pk=5.1, x=0.1	0

TABLE NO 2 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
136	m1	-	-		3.2		
	m2	0	(-)	0	3.1		
	m3	(-)	(-)		3.3		
	m4	0	0	(-)	3.4		
137	m1	(-)	(-)		3.0		
	m2	(+)	(-)	0	2.7		
	m3	0	(-)		3.6		
	m4	(+)	(-)	(-)	3.1		
158	m1	(-)	(-)				
	m2	(-)	(-)	(-)			
	m3	(-)	(0)				
	m4	(-)	(-)	(-)			
159	m1	(-)	(-)				
	m2	(-)	(-)	(-)			
	m3	(-)	(-)				
	m4	(-)	(-)	(-)			
164	m1	--	--		4.5		
	m2	-	-	-	4.4		
	m3	-	-		4.0		
	m4	-	-	-	4.3		
165	m1	--	--		3.7		
	m2	--	--	--			
	m3	--	--		4.4		
	m4	--	--	--	4.2		
168	m1	--	--		5.0		
	m2	+	-	-	4.7		
	m3	--	-		4.3		
	m4	-	-	-	4.8		
169	m1	(+)	(-)	(-)			
	m2	(+)	(-)	(-)	(-)		
	m3	0	0		(3.9)		
	m4	(+)	0	0	(-)		
170	m1	(+)	(-)		3.8		
	m2	(+)	(-)	0	(3.5)		
	m3	+	0		4.2		
	m4	+	(-)	(-)	4.1		
171	m1	(+)	(-)		0		
	m2	(+)	(-)	0	0		
	m3	0	0		0		
	m4	(+)	0	0	0		

TABLE NO. 2 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
172	m1	0	0				
	m2	0	0	0			
	m3	0	0				
	m4	(-)	(-)	(-)			
173	m1	(+)	(+)		0		
	m2	0	(-)	0	0		
	m3	0	+		0		
	m4	(-)	0	(-)	0		
174	m1	0	0				
	m2	-	-	0			
	m3	0	0				
	m4	(-)	(-)	(-)			
182	m1	-	-		(4.7)		
	m2	-	-	-	(4.5)		
	m3	(-)	(-)		(4.2)		
	m4	-	-	-	(4.5)		
183	m1	--	--		4.4		
	m2	--	--	--	4.8		
	m3	--	--		3.8		
	m4	--	--	--	4.6		
184	m1	--	--		4.0		
	m2	--	--	--	4.0		
	m3	--	--		3.6		
	m4	--	--	--	4.1		
185	m1	0	(+)		(4.3)		
	m2	+	-	-	(4.6)		
	m3	-	-		(4.4)		
	m4	+	0	-	(4.5)		
193	m1	-	-		<5		
	m2	(-)	0	0	<5		
	m3	-	-		<5		
	m4	-	-	-	<5		
201	m1	--	--				
	m2	-	--	-			
	m3	--	--				
	m4	-	-	-			
202	m1	--	--				
	m2	-	--	--			
	m3	--	--				
	m4	--	--	--			

TABLE NO. 2 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
203	m1	--	--				
	m2	-	--	--			
	m3	--	--				
	m4	--	--	--			
204	m1	--	--		4.4		
	m2	-	-	-	(4.7)		
	m3	--	--		4.2		
	m4	--	--	--	4.6		
205	m1	--	--		4.9		
	m2	+	-	-	5.2		
	m3	-	-		4.3		
	m4	-	-	--	4.3		
206	m1	--	--		4.2		
	m2	-	-	-	4.0		
	m3	-	-		(3.9) (4.3)		
	m4	--	--	--	4.3		
207	m1	--	--		-		
	m2	--	--	--	-		
	m3	--	--		-		
	m4	--	--	--	-		
246	m1	--	--		3.6		
	m2	--	--	--	3.7		
	m3	--	--		3.7		
	m4	--	--	--	4.0		
247	m1	+	0		(4.7)		
	m2	+	(-)	(-)	(4.4)		
	m3	(-)	(-)		(4.3)		
	m4	++	-	-	(5.6)		
248	m1	(+)	++		(4.6)		
	m2	0	++	0	(4.5)		
	m3	0	++		(4.6)		
	M4	0	++	-	(4.6)		
249	m1	-	++		(4.6)		
	m2	-	++	-	(4.2)		
	m3	-	++		(4.6)		
	m4	-	++	-	(4.4)		
404	m1	+	-		5.2		
	m2	+	+	0	5.7		
	m3	+	-		5.2		
	m4	+	+	-	5.9		

TABLE NO. 2a

Compound no.	Receptor	SECONDARY ASSAY pK	xNMS	xACh
101	m1	7.2	0.007	0
	m2			
	m3			
	m4			
106	m1			
	m2			
	m3	4.0	3.8	3.9
	m4			
121	m1	7.4	0	0
	m2			
	m3			
	m4			
123	m1	3.6	3	0
	m2			
	m3			
	m4			
124	m1	5.1	1.2	0.4
	m2			
	m3			
	m4			
134	m1	3.5	0	4.7
	m2			
	m3			
	m4			
135	m1	5.6	0.9	0.2
	m2	5.2	0.6 with ^{3}H-P??	4.9
	m3	5.1	1.7	0.3
	m4			
173	m1	I		
	m2			
	m3	I		
	m4			

TABLE NO. 3

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
211	m1	-	-				
	m2	-	-	-			
	m3	(-)	(-)				
	m4	-	-	-			
212	m1	-	-				
	m2	-	-	-			
	m3	-	-				
	m4	-	-	-			
222	m1	-	-				
	m2	-	-	-			
	m3	-	-				
	m4	-	-	-			
223	m1	(+)	-		(4.6)		
	m2	+	-	-	(4.4)		
	m3	-	-		(3.7)		
	m4	0	-	-	5.7		
224	m1	+	-		5.5		
	m2	0	-	-	5.5		
	m3	+	-		5.7		
	m4	0	-	-	5.7		
225	m1	-	-		4.9		
	m2	-	-	-	5.0		
	m3	+	(-)		5.3		
	m4	0	-	-	5.5		
226	m1	(+)	-		(5.3		
	m2	+	-	-	(5.2)		
	m3	+	-		(5.5)		
	m4	+	-	-	(5.5)		
227	m1	-	-		(4.6)		
	m2	-	-	-	(4.8)		
	m3	-	+		(4.4)	0	
	m4	-	-	-	(4.5)		
228	m1	-	-		(5.0)		
	m2	(+)	-	-	(5.4)		
	m3	-	-		(4.6)		
	m4	-	-	-	(5.1)		
229	m1	-	-		5.2		
	m2	-	-	-	4.5		
	m3	-	-		4.3		
	m4	-	-	-	5.0		

TABLE NO. 3 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
307	m1	(+)	-		(5.4)		
	m2	+	-	-	(5.2)		
	m3	+	-		(5.4)		
	m4	+	-	-	(5.5)		
308	m1	-	-		(4.6)		
	m2	-	-	-	(5.0)		
	m3	-	-		(4.5)		
	m4	-	(+) -	-	(4.7)		
314	m1	-	-	-			
	m2	-	-	(+) -	-	0	
	m3	(-)	(-)		-		
	m4	-	(+) -	-	-		
315	m1	-	-	-			
	m2	-	(+) -	-	-	0	
	m3	-	-	-			
	m4	-	-	-	-		
316	m1	-	-		(4.5)		
	m2	-	-	-	(4.8)		
	m3	-	0		(4.2)		
	m4	-	-	-	(4.4)		
317	m1	--	--		4.4		
	m2	(-)	--	--	4.6		
	m3	-	-		3.6		
	m4	-	-	--	(4.5)		
322	m1	-	-		(4.5)		
	m2	0	-	-	(4.9)		
	m3	-	-		(4.4)		
	m4	-	-	-	(4.7)		
325	m1	--	--		6.0		
	m2	--	--	--	6.2		
	m3	--	--		(5.5)		
	m4	--	--	--	6.0		
330	m1	-	-		(5.2)		
	m2	-	-	-	(5.4)		
	m3	-	-		(5.3)		
	m4	-	-	-	(5.2)		
331	m1	-	-		0		
	m2	-	-	-	0		
	m3	-	-	-			
	m4	-	-	-	0		

TABLE NO. 3 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
333	m1	-	-		5.3		
	m2	(-)	-	-	5.1		
	m3	(+)	-		5.3		
	m4	(+) -	-	-	5.4		
334	m1	-	-				
	m2	-	-	-			
	m3	-	-				
	m4	-	-	-			
335	m1	-	-		4.5		
	m2	(-)	-	-	4.8		
	m3	-	-		(3.7)		
	m4	-	-	-	4.3		
344	m1	(+) -	-		5.5		
	m2	+-	-	-	5.6		
	m3	-	-		4.9		
	m4	+-	-	-	5.3		
381	m1	0	-		(4.7)		
	m2	(-)	-	-	(4.2		
	m3	-	-		(6.3) max = 30		
	m4	+	-	-	(4.7)		
382	m1	(-)	(-)				
	m2	(-)	(-)	(-)			
	m3	-	-				
	m4	(-)	-	-			
390	m1	-	--		4.5		
	m2	0	--	--	(4.2)		
	m3	-	-		(3.5)		
	m4	-	--	--	(3.9)		
391	m1	--	--		5.7		
	m2	+	--	--	5.6		
	m3	--	--		4.8		
	m4	+	--	--	5.5		
394	m1	-	-		4.7		
	m2	+	--	--	5.6		
	m3	-	-		4.4		
	m4	+	--	--	5.1		
396	m1	+	--		5.1		
	m2	+	--	--	5.3		
	m3	+	-		4.7		
	m4	+	--	--	5.1		

TABLE NO. 3 contd.

Compound no.	Receptor	^{3}H-NMS	ACh	^{3}H-ACh	Off-rate	GTP	SMB
397	m1	--	--		5.1		
	m2	+	--	--	6.0		
	m3	-	--		4.5		
	m4	(--)	--	--	5.4		
398	m1	-	-		4.5		
	m2	(-)	-	-	4.3		
	m3	-	-		(3.8)		
	m4	0	-	-	4.4		
402	m1	+	--		6.1		
	m2	+	--	--	6.0	pk=6.1±.02 α=0.003±.002	
	m3	0(--)	--		5.2		
	m4	+	--	--	6.0		
403	m1	-	--		5.7		
	m2	+	--	--	6.1		
	m3	-	-		4.8		
	m4	+	-	-	5.7		
405	m1	-	--				
	m2	-	--	--			
	m3	-	-				
	m4	-	-	-			
438	m1	--	--				
	m2	--	--	--	0		
	m3	--	--				
	m4	--	--	--			
439	m1	-	-				
	m2	-	-	-			
	m3	-	-				
	m4	-	-	-			

TABLE NO. 3a

Compound no.	Receptor	SECONDARY ASSAY pK	αNMS	αACh
227	m1	5.7	0.1	0.3
	m2			
	m3	5.2	0.4	1.2
	m4			
228	m1	5.2	0.1	0.6
	m2			
	m3			
	m4			

M&C FOLIO: 230P70859/FP-9414 WANGDOC: 1011D

Claims

1. A method of regulating receptor response in vivo in a mammalian subject, said receptor being selected from m1, m2, m3, m4, m5, M1, M2, M3, M4 and M5 muscarinic receptors, comprising the step of administering to said subject an effective amount of a selective allosteric effector to regulate said receptor.

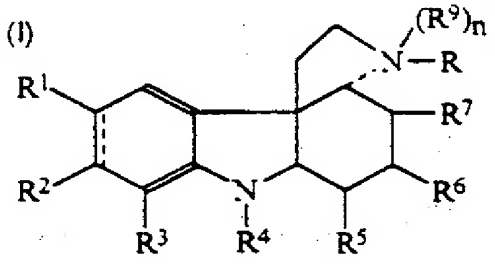
2. A method according to claim 1, wherein the receptors for regulation are the m1, m2, m3 and m4 receptors.

3. A method according to claim 1, wherein the receptor for regulation is the m1 receptor.

4. A method according to any preceding claim, wherein the effector exhibits positive cooperativity with acetylcholine at the receptor.

5. A compound of formula (I):

Formula (I)



(I)

[wherein the dashed lines independently indicate the presence or absence of a carbon-carbon bond;

R^1 and R^2 are the same or different and, where the dashed line indicates a carbon-carbon bond, may each represent a hydrogen atom, a hydroxy group, a lower

alkoxy group, an amino group, an amino group substituted with one or two lower alkyl groups, a nitroso group, a nitro group, a carboxyl group, a carbamoyl group or a carbamoyl group substituted with one or two lower alkyl groups,

or, where the dashed line indicates no bond, then one of R^1 and R^2 represents a keto group and the other represents a keto group or two hydrogen atoms;

R^3 represents a hydrogen atom, a hydroxy group or a lower alkoxy group;

R^4 represents a hydrogen atom or an amino-protecting group, such as a lower carboxylic acyl group;

R^5 represents a hydrogen atom or a lower carboxylic acyl group;

R^6 represents a hydrogen atom;

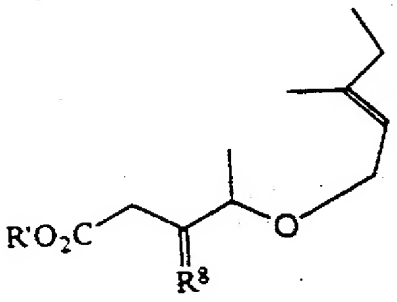
R^7 represents a hydrogen atom or, together with R, represents a lower alkylene group;

or

R, together with R^6, represents a lower alkylene group substituted with a hydroxy-loweralkenyl group;

or

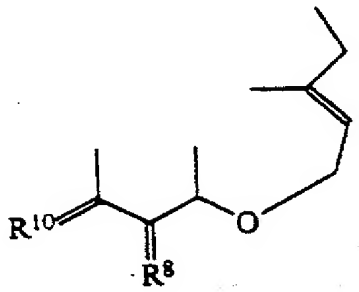
R, together with R^5 and R^6, represents a group of formula:



in which R^8 represents a hydroxyimino group or two hydrogen atoms, and R' represents a carboxy-protecting group;

or

R, together with R^4, R^5 and R^6, represents a group of formula (i):



(i)

in which R^8 is as defined above and R^{10} represents a keto group or two hydrogen atoms;

R^9 represents a hydroxy group, an amino group, an amino group substituted with one or two lower alkyl groups, an aryl-carboxylic acyl group optionally substituted with one or more substituents selected from Substituents (a) below, or an alkyl group, an alkenyl group or alkynyl group which is straight or branched and which is optionally substituted with a substituent selected from:

sulfonyl groups, carboxyl groups, cyano groups, cycloalkyl groups, heteroaryl groups having from 5 to 10 ring atoms, 1 to 3 of which are selected from O, S and N, aryl and biaryl groups having from 1 to 13 carbon atoms and which may be further substituted with one or more substituents selected from Substituents (a) below;

n=0 or 1;

Substituents (a): halo, hydroxy, amino, nitro, azido and cyano groups;

or,

when the compound of formula (I) is a Wieland-Gumlich aldehyde (in which R^4 represents a hydrogen atom and R^5 represents a CHO group), a dimer thereof wherein the substituent R^4 from each monomer forms, together with the substituent R^5 from the other monomer, a methenylene link;

and pharmaceutically acceptable salts and esters thereof]

in therapy.

6. A compound according to claim 5 in the form of a Wieland-Gumlich aldehyde and wherein R^7 is a hydrogen atom.

7. A compound according to claim 5 or 6, wherein the group represented by R^6 and R is an ethylene group and the hydroxy-loweralkenyl group is 2-hydroxy-1-ethenyl spaced apart from R^5 by two carbon atoms.

8. A compound according to any of claims 5 to 7, wherein R^4, R^5, R^6 and R represent a group of the formula (i).

9. A compound according to claim 8, wherein R^8 represents two hydrogen atoms and R^{10} represents a keto group.

10. A compound according to any of claims 5 to 9, wherein the dashed lines both represent carbon-carbon bonds.

11. A compound according to any of claims 5 to 10, wherein any lower alkyl, lower alkenyl, lower alkoxy, lower alkylene and lower alkenylene have from 1 to 4

carbon atoms.

12. A compound according to claim 11, wherein any lower alkyl, lower alkenyl, lower alkoxy, lower alkylene and lower alkenylene have from 1 to 3 carbon atoms.

13. A compound according to any of claims 5 to 12, wherein any lower alkyl groups are ethyl or methyl groups.

14. A compound according to any of claims 5 to 12, wherein any lower alkyl groups are methyl groups.

15. A compound according to any of claims 5 to 12, wherein any lower alkoxy groups are methoxy groups.

16. A compound according to any of claims 5 to 15, wherein any alkyl, akenyl or alkynyl group not specified as being "lower" has from 1 to 10 carbon atoms.

17. A compound according to claim 16, wherein any alkyl, akenyl or alkynyl group not specified as being "lower" has from 1 to 6 carbon atoms.

18. A compound according to any of claims 5 to 17, wherein R^2 is lower-alkoxy, hydrogen or hydroxy. When R^2 is lower-alkoxy, then we prefer that it should be methoxy.

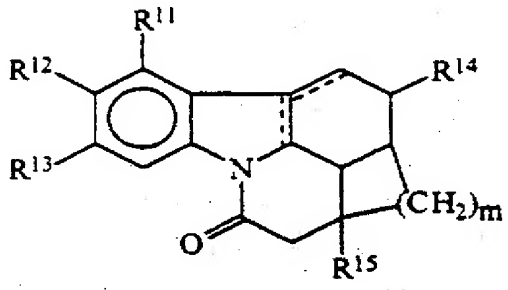
19. A compound according to claim 18, wherein when R^2 is lower-alkoxy then it is methoxy.

20. A compound according to any of claims 5 to 19, wherein R^1 is as defined for R^2 in either of claims 18 or 19, and may be the same or different.

21. A compound according to claim 20, wherein R^1 and

R^2 are the same and are both hydrogen or methoxy.

22. The use of compounds of formula (II):



(II)

[wherein the dashed lines optionally represent an additional carbon-carbon bond, provided that both do not represent additional carbon-carbon bonds;

R^{11}, R^{12} and R^{13} are the same or different and each represents a hydrogen atom, a hydroxy group, a carbamoyl group, a carbamoyl group substituted with one or two lower alkyl groups, a lower-alkoxy group, an aralkoxy group in which the aryl part has from 6 to 10 carbon atoms and the alkyl is a lower alkyl or an aryloxy group in which the aryl group has from 6 to 10 carbon atoms;

R^{14} represents a carboxy group, a carbamoyl group, a carbamoyl group substituted with one or two lower alkyl groups, or R^{14} represents a group of formula $-NR^{16}R^{17}$ wherein R^{16} and R^{17} are the same or different and each represents a hydrogen atom, a hydroxyalkyl group, a lower alkyl group optionally substituted by one or more substituents (a) as defined in claim 5, a carboxy-loweralkyl group, or R^{16} and R^{17} together represent an akylene group having from 3 to 6 carbon atoms and which may be substituted by substituents (a) as defined in claim 5, or one of R^{16} and R^{17} represents a hydrogen atom and the other

represents an imino-loweralkyl group, a lower-alkoxy group, an aralkoxy group in which the aryl part has from 6 to 10 carbon atoms and the alkyl part is a lower alkyl, an aryloxy group in which the aryl group has from 6 to 10 carbon atoms, the above aryl parts being optionally substituted with one or more substituents selected from substituents (a) as defined in claim 5,

R^{15} represents a hydrogen atom, a lower alkyl group, an aryl group having from 6 to 10 carbon atoms, or an aralkyl group in which the aryl part has from 6 to 10 carbon atoms and the alkyl part is a lower alkyl;

and salts and esters thereof, including thioesters]

in therapy.

23. A compound according to claim 22, wherein m is 2, 3 or 4, and is particularly preferably 3.

24. A compound according to claim 22, wherein m is 3.

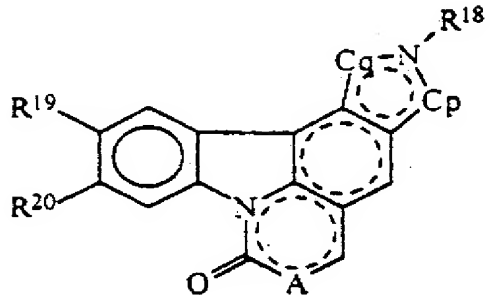
25. A compound according to any of claims 22 to 24, wherein R^{15} is a hydrogen atom.

26. A compound according to any of claims 22 to 25, wherein R^{11} represents a hydrogen atom or a hydroxy group.

27. A compound according to any of claims 22 to 26, wherein R^{12} and R^{13} both represent hydrogen atoms.

28. A compound according to any of claims 22 to 26, wherein one of R^{12} and R^{13} represents a hydroxy or a methoxy group and the other represents a hydrogen atom or a hydroxy or methoxy group respectively.

29. The use of compounds of formula (III):



(III)

[wherein p+q=2, 3, or 4 or C_q-N-R^{18} is only attached to the remainder of the molecule by C_q and p=0;

rings containing dashed lines are saturated, partially unsaturated or completely unsaturated;

R^{18} represents a hydrogen atom, a lower-alkoxycarbonyl group or an aralkoxy group in which the aryl part has from 6 to 10 carbon atoms and the alkyl part is a lower alkyl and the aryl part is optionally substituted with one or more substituents selected from substituents (a) as defined in claim 5;

R^{19} and R^{20} are the same or different and each represents a hydrogen atom, a lower alkoxy group or a hydroxy group;

A represents =CH-, $-CH_2-$, -O- or -NH-;

and salts and esters thereof]

in therapy.

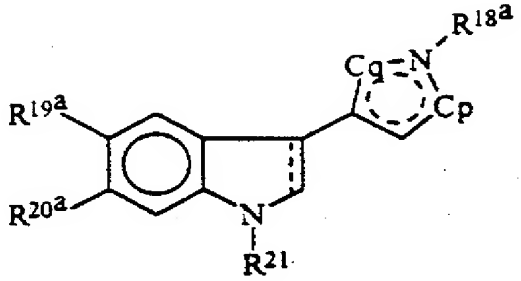
30. A compound according to claim 29, wherein p+q=3.

31. A compound according to claim 29 or 30, wherein R^{19} and R^{20} are as defined for for R^1 and R^2 and

R^{12} and R^{13} in any preceding claim.

32. A compound according to any of claims 29 to 31, wherein A represents =CH- or $-CH_2-$.

33. The use of compounds of formula (IV):



(IV)

[wherein the dashed ring indicates unsaturation, saturation, or partial unsaturation, and the dashed line represents an optional extra carbon-carbon bond;

R^{18a}, R^{19a} and R^{20a} have the same meanings as for R^{18}, R^{19} and R^{20} in any preceding claim, respectively;

R^{21} represents a hydrogen atom or a group of formula $-C(O)_r-A'-CH_2-Z$ in which r=0 or 1, A' is $-CH_2-$, -O- or -NH- and Z is a vinyl or ethynyl group;

and salts and esters thereof]

in therapy.

34. A compound according to claim 33, wherein the substituents are as defined for the substituents in any of laims 29 to 32.

35. A compound according to claim 33 or 34, wherein, in the substituents R^{21}, r=1.

36. The use of a compound according to any preceding claim in the manufacture of a medicament for the treatment of a condition responsive to allosteric stimulation of muscarinic receptors.

37. Use according to claim 36 wherein the condition is dementia.

38. Use according to claim 36 wherein the condition is senile dementia.

39. Use according to claim 36 wherein the condition is Alzheimer's Disease.

40. A compound according to any of claims 1 to 35, for use as sedatives for the CNS.

41. The use of a compound according to any of claims 1 to 35 in the treatment of the following conditions:

Alzheimer's Disease;
Parkinson's Disease;
Motion sickness;
Huntingdon's chorea;
Schizophrenia;
Depression;
Anxiety;
Sedation;
Analgesia;
Stroke;
Preanaesthetic;
Antispasmodic;
Irritable Bowel Syndrome;
Bladder - incontinence, retention;
Peptic ulcer disease;
Bronchitis/asthma/cronic obstructive airways disease;
Sinus bradycardia;
Pacemaker regulation;

Glaucoma;
Achalasia;
Symptomatic diffuse oesophageal spasm;
Biliary dyskinesia;
Scleroderma;
Diabetes mellitus;
Lower oesophageal incompetence;
Intestinal pseudo obstruction;
Regulation of sleep;
Control of pupil diameter; and/or
Non-ulcer dyspepsia.

Patents Act 1977 223
Examiner's report to the Comptroller under Section 17 (The Search report)

Application number
GB 9415174.3

Relevant Technical Fields

(i) UK Cl (Ed.N) A5B: BHA, BJA, BJB

(ii) Int Cl (Ed.6) A61K

Search Examiner
MRS S E CHALMERS

Date of completion of Search
6 NOVEMBER 1995

Databases (see below)
(i) UK Patent Office collections of GB, EP, WO and US patent specifications.

(ii) ONLINE: WPI, CLAMS, CHEMICAL ABSTRACTS

Documents considered relevant following a search in respect of Claims :-
1-4; 36-41 WHEN APPENDENT TO CLAIMS 1-4

Categories of documents

X: Document indicating lack of novelty or of inventive step.

Y: Document indicating lack of inventive step if combined with one or more other documents of the same category.

A: Document indicating technological background and/or state of the art.

P: Document published on or after the declared priority date but before the filing date of the present application.

E: Patent document published on or after, but with priority date earlier than, the filing date of the present application.

&: Member of the same patent family; corresponding document.

Category	Identity of document and relevant passages	Relevant to claim(s)
X	Life Sciences, vol 55 (25/26) 2135-2145, (1994) F J Ehlert et al "Muscarinic Receptors and Novel Strategies for the Treatment of Age-Related Brain Disorders" (see especially page 2142)	1 at least
X	J Clinic Invest, vol 91, 1314-1318 (1993) D B Jacoby et al "Human Eosinophil Major Basic Protein is an Endogenous Allosteric Antagonist at the Inhibitory Miscarinic M2 receptor"	1 at least
X	J Pharmacol Exp Ther, vol 258(3) 992-998 (1991) Endou et al "Binding Profiles of Class I Anti-arrhythmic Agents to Cardiac Muscannic Receptors; Competitive and Allosteric Interactions and their Pharmacological Significance"	1 at least

Databases:The UK Patent Office database comprises classified collections of GB, EP, WO and US patent specifications as outlined periodically in the Official Journal (Patents). The on-line databases considered for search are also listed periodically in the Official Journal (Patents).

Continuation page

Category	Identity of document and relevant passages	Relevant to claim(s)
X	Mol Pharmacol, vol 38(5) 674-680 (1990) Tucek et al "Positive co-operativity in the binding of Alcuronium to Muscarinic Acetylcholine Receptors"	1, 4
X	Br J Pharmacol, vol 98 79-86 (1991) Hunter et al "The cholinergic pharmacology of tetrahydroaminoacridine in vivo and in vitro"	1 at least
X	J Cardiovasc Pharmacol, vol 11(2) 222-229 (1988) Brumner et al "Binding of 2 specific bradycardic agents, alinidine and AQ-A 39 to muscannic receptors of guinea pig atria and ventricle"	1 at least